WHO monographs on

selected medicinal plants

Volume 4

World Health Organization

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VOLUME 4

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WHO also acknowledges with thanks the valuable work of the approximately 200 experts including 81 national health authorities, who provided comments and advice on the draft texts; those who submitted comments through the World Self-Medication Industry (a nongovernmental organization in official relations with WHO) and the International Federation of Pharmacists (a nongovernmental organization in official relations with WHO); and those who participated in the Fourth WHO Consultation on Selected Medicinal Plants held in Salerno-Paestum, Italy, in October 2005 to review the monographs (see Annex 1).

Sincere appreciation is extended to the Ministry of Health of Italy, the Government of the Province of Campagna, Italy, the Municipal Government of Salerno, Italy, and the State University of Salerno, who hosted the above-mentioned Fourth WHO Consultation and supported it financially. Finally, WHO wishes to express thanks to Mr Raymond Tsai, Boston, USA, Dr Hermann Garden, Basel, Switzerland, Ms Lynn Morra, Abu Dhabi, United Arab Emirates, and Ms Tina Lu, Rochester, USA, for their indispensable assistance in finalizing and editing the manuscripts.

Introduction

Increasing role of the *WHO monographs on selected medicinal plants*

Over the past two decades, there has been a tremendous increase in the use of herbal medicine; however, there is still a significant lack of research data in this field. Therefore since 1999, WHO has published three volumes of the *WHO monographs on selected medicinal plants*: volume 1 includes 28 monographs; volume 2 contains an additional 30 monographs; and volume 3 provides 31 monographs. Including the 28 new monographs published in this volume, a total of 118 monographs in four volumes are now available on the WHO web site (http://www.who.int/medicinedocs/en/m/abstract/Js14213e/).

Due to the diversity of medicinal plants and herbal medicines, it is difficult for WHO to continue to develop more monographs on commonly used medicinal plants. One of the objectives of WHO monographs is to provide a model that will support countries in developing their own national or regional monographs on medicinal plants or national formularies on herbal medicines. Experts can be trained through the process of developing country-specific or regional monographs, and national capacity in this field can thus be built up.

For example, at WHO's regional training workshop on regulation of herbal medicines held for the WHO European Region, in September 2003, the participating national drug regulatory authorities of many of the Newly Independent States (NIS) submitted their request to WHO directly, for assistance in the development of monographs on medicinal plants commonly used in NIS.

In order to respond to their urgent need, WHO initiated a new project to develop a set of regional (NIS) monographs on commonly used medicinal plants, based on available scientific information relating to their safety, efficacy and quality, which will facilitate the creation of effective and practical regulatory and quality assurance measures on herbal medicines. WHO has been working with 15 national drug regulatory authorities interested in this publication in NIS, Countries of Central and Eastern

Europe (CCEE) and their neighbouring countries, in close collaboration with the WHO Regional Office for Europe. The 13 new monographs on commonly used medicinal plants in NIS have been drafted based on the format of the WHO monographs by the experts in NIS and CCEE countries with the support of experts, national health authorities and NGOs within and also outside the NIS and CCEE countries. The *WHO monographs on medicinal plants commonly used in NIS* have been completed and will be published soon. Based on the NIS countries model, in the future, WHO would like to cooperate with more countries or regions to develop their monographs on commonly used medicinal plants.

Preparation of monographs for volume 4

Selection of medicinal plants

The selection of medicinal plants for inclusion in the WHO monographs is based on worldwide use. The medicinal plants selected must meet two major criteria: (1) they must be in common use in at least two WHO Regions; and (2) there must be sufficient scientific data available to satisfy the requirements of the various sections in the monograph format.

The recommended selection criteria discussed at the Third WHO Consultation on Selected Medicinal Plants (Ottawa, Canada, July 2001) were applied to the preparation of volume 4 of the WHO monographs.

Preparation

During the preparation of volume 4, more than 200 experts were involved in addition to members of WHO's Expert Advisory Panel on Traditional Medicine, a significant expansion compared to the numbers involved in the previous three volumes. National drug regulatory authorities in 81 countries participated in the process, again a greater number than for the previous volumes. This global network of active players facilitated wider access to the available scientific references and information, in terms of both quality and quantity. This considerable level of support contributed greatly to the efficiency of the preparation process.

The Fourth WHO Consultation on Selected Medicinal Plants was held in Salerno-Paestum, Italy, in October 2005 to review and finalize the draft monographs. Thirty-four experts and drug regulatory authorities from WHO Member States participated (Annex 1). Following extensive discussion, 28 of the 32 draft monographs were adopted for inclusion.

Changes in format in volume 4

A description of selected sections of the monographs is given in the General technical notices, which reflect the above-mentioned format

changes. For easy reference, two cumulative indexes are provided as annexes. Annex 2 lists the monographs in alphabetical order of the plant name, while Annex 3 is arranged according to the plant materials of interest. For the convenience of readers, an additional cumulative list of plant names (scientific name followed by family name) which appeared under the heading "definition" in each monograph, has been added (Annex 4). In response to the recommendation of the Ottawa Consultation in 2001, a cumulative index of major chemical constituents arranged in alphabetical order (Annex 5), a cumulative index ordered by Chemical Abstracts Service (CAS) number (Annex 6) and a cumulative index ordered by molecular formula (Annex 7) have also been included in this volume.

Under the heading "Geographical distribution", an attempt has been made to describe the geographical distribution of the plant, i.e. its natural distribution, where it is cultivated, and conditions of cultivation, harvesting and storage. This has been a challenge, owing to the lack of data based on established national good agricultural practices and/or good collection practices (GACP) for medicinal plants. In 2007, WHO published the *WHO guidelines on good manufacturing practices (GMP) for herbal medicines*, which provide general technical guidance on obtaining medicinal plant materials of good quality for the production of herbal medicines in the overall context of quality assurance and control of herbal medicines. WHO also compiled new guidelines on assessing quality for safety of herbal medicines with reference to contaminants and residues which were published in 2007. It is hoped that these guidelines will facilitate the implementation of GMP for herbal medicines at the national level, and the development of national quality standards/specifications for herbal medicines, which in turn should bridge the current information gap in this area.

At the consultation, the following technical issues were pointed out for consideration during the preparation of the monographs:

- Specify extract according to original literature, whenever possible.
- Posology: as a general rule make clear that a given posology has been found either in the report of clinical trials or referring to traditional indications. These guidelines have been followed in the preparation of this volume wherever possible.
- Provide posologies as they relate to the specific indications – the dosage forms have to be consistent with their respective indications.
- Specific posologies should be indicated for specific uses and dosage forms whenever available.

Purpose and content of monographs

Although the monographs include one section on "medicinal uses" with three categories of information, the purpose of the monographs was clearly explained in the introduction to volume 1, and it is unnecessary to repeat it here. But it should be noted that the word "monograph" is used as a technical term only. It does not have the same meaning as "monograph" in any type of pharmacopoeia. In addition, it is reiterated here that this publication is not intended to replace any official compendia such as pharmacopoeias, formularies or legislative documents.

It should also be emphasized that the descriptions included in the section on medicinal uses should not be taken as implying WHO's official endorsement or approval. They merely represent the systematic collection of scientific information available at the time of preparation, for the purpose of information exchange.

Finally, the Fourth WHO Consultation on selected medicinal plants recommended WHO to update existing monographs in view of the availability of new data and of the format change that has been employed for volumes 3 and 4, and that this process should be given priority over the development of new monographs, if resources are limited. This volume might therefore be the last volume of WHO monographs on commonly used medicinal plants. We should like to express our appreciation to all the experts, national health authorities, WHO collaborating centres and NGOs for their efforts, technical contributions and support in the preparation of the four volumes of WHO monographs on selected medicinal plants.

In the future, WHO will consider updates of existing volumes of monographs and will focus on providing technical assistance for national capacity building through development of sets of regional and/or subregional monographs by transferring the know-how and by mobilizing the established global network.

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General technical notices

These WHO monographs are not pharmacopoeial monographs. Their purpose is to provide scientific information on the safety, efficacy and quality control/quality assurance of widely used medicinal plants, in order to facilitate their appropriate use in WHO's Member States; to provide models to assist WHO's Member States in developing their own monographs or formularies for these and other herbal medicines; and to facilitate information exchange among WHO's Member States.

The format used for volume 4 follows that of volume 3, which was essentially that of volume 2, with the following modification made: to keep relevant sections together, *Adverse reactions* appears immediately after the section on *Pharmacology*. The titles of three categories under the *Medicinal uses* have been changed to the following:

- *Uses supported by clinical data*
- *Uses described in pharmacopoeias and well established documents*
- *Uses described in traditional medicine*

The *Definition* provides the Latin binomial name, the most important criterion in quality assurance. Latin binomial synonyms and vernacular names, listed in *Synonyms* and *Selected vernacular names* respectively, are names used in commerce or by local consumers. The monographs place outdated botanical nomenclature in the synonyms category, based on the International Code of Botanical Nomenclature. The vernacular names comprise an alphabetical list of selected names from individual countries worldwide, in particular from areas where the medicinal plant is in common use. They refer to the medicinal plant itself not the medicinal plant part, which is identical to the monograph name. The lists are not complete, but reflect the names of the concerned medicinal plant appearing in the official monographs and reference books consulted and those in the Natural Products Alert (NAPRALERT) database (a database of literature from around the world on ethnomedical, biological and chemical information on medicinal plants, fungi and marine organisms, located at the WHO Collaborating Centre for Traditional Medicine at the University of Illinois at Chicago, Chicago, IL, USA). While every effort has been made to delete names referring to the

medicinal plant part, the relevant section of each monograph may still include these.

Geographical distribution is not normally found in official compendia, but is included here to provide additional quality assurance information. The detailed botanical description under *Description* is intended for quality assurance at the stages of production and collection; the description of the crude drug material under *Plant material of interest* is for the same purpose at the manufacturing and commerce stages.

General identity tests, *Purity tests* and *Chemical assays* are all normal compendial components included under those headings in these monographs. Where purity tests do not specify accepted limits, those limits should be set in accordance with national requirements by the appropriate authorities of Member States.

Each medicinal plant and the specific plant part used as crude drug material contains active or major chemical constituents with a characteristic profile that can be used for chemical quality control and quality assurance. These constituents are described in the *Major chemical constituents*.

Descriptions included in *Medicinal uses* should not be taken as implying WHO's official endorsement or approval for such uses. They merely represent the systematic collection of scientific information available at the time of preparation, for information exchange.

The first category, *Uses supported by clinical data*, includes medical indications that are well established in some countries and have been validated by clinical studies documented in the scientific literature. Clinical trials may be controlled, randomized, double-blind studies, open trials, cohort studies or well documented observations on therapeutic applications.

The second category, *Uses described in pharmacopoeias and well established documents*, includes medicinal uses that are well established in many countries and are included in official pharmacopoeias or governmental monographs. Uses having a pharmacologically plausible basis are also included, as well as information resulting from clinical studies that clearly need to be repeated because of conflicting results.

The third category, *Uses described in traditional medicine*, refers to indications described in unofficial pharmacopoeias and other literature, and to traditional uses. Their appropriateness could not be assessed, because sufficient data to support the claims could not be found in the literature. Traditional uses that address severe pathologies, such as cancer, AIDS, hepatitis, etc., as they relate to these modern biomedical terms, should only be included under the third heading if pharmacological data or robust ethnopharmacological/ethnobotanical reports are available to support the claims.

The *Experimental pharmacology* section includes only the results of investigations that prove or disprove the cited medicinal uses. Abbreviated details of the best-performed studies have been included in this section. Other published experimental data that are not associated with the medicinal uses have not been included, to avoid confusion.

The details included in the *References* have been checked against the original sources wherever possible. For references in languages other than English, except for those in Chinese, Japanese and Korean, the title is given in the original language, except in cases where an English summary is available.

Fructus Agni Casti

Definition
Fructus Agni Casti consists of the dried, ripe fruits of *Vitex agnus-castus* L. (Lamiaceae) (*1*, *2*).

Synonyms
Agnus-castus vulgaris Carr., *Vitex verticillata* Lam. (*3*).

Selected vernacular names
Abraham's balm, Abrahamsstrauch, agneau-chaste, agnocasto, agnos-casto cumune, agnus-castus, angarf, ârbol casto, ârbolde la castidad, arbre au poivre, athlak, banjankusht, barátcserje, bish barmagh aghaji, chaste tree, chasteberry, common chaste tree, daribrahim, felfele barry, fanfangosht, gatileira comum, gattilier, gattilier commun, hab an nasl, hab el fakd, hab a khouraf, hayit, hemp tree, jurema, kaff maryam, kef-meriem, kerwa, Keuschbaum, Keuschlamm, kyskhedstrae, lilac chastetree, lygos, Mönchspfeffer, Mönchspfeller, monk's pepper, monk's pepper tree, Müllen, non's peppertree, panj angosht, panjangusht, pape falso, peperella, petite poivre, pimiento menor, poivre de moine, poivre sauvage, ranukabija mah, sagetree, sauzgatillo, seiyo-ninzin-boku, shajerat Ebrahim, shagareh Ibrahim, sinduvara, tree of chastity, true chaste tree, vitex, vitiu, wild lavender, Yemen safrani (*1–7*).

Geographical distribution
Native to the Mediterranean region and Asia (*2*, *4*, *8*). Cultivated in warm temperate regions of the world, and obtained primarily from Mediterranean countries, especially Albania and Morocco (*3*, *9*).

Description
A small tree or deciduous shrub, approximately 1–6 m in height, with aromatic odour. Leaves: opposite, long-petiolate, palmately-compound with 3–9 stipulate leaflets; leaflet blade linear-lanceolate, apex and base acuminate, 1.5–10.0 cm long, 0.5–2.0 cm wide; the central leaflet is the longest,

dark green and glabrous above, velvety white-tomentose below; margin entire to sparsely toothed. Inflorescence: terminal panicle, 12.0–17.5 cm long, and composed of many sessile-subsessile cymes. Flower: perfect, campanulate symmetric, white-tomentose; calyx 5-toothed, campanulate, 2.0–2.5 cm long; corolla blue, pink, yellowish or white, salverform, tube 6–7 mm long, limb 2-lipped, upper lip 2-lobed, lower lip 3-lobed; stamens 4, exerted, 2 long, 2 short, inserted near top of corolla tube, alternate with corolla lobes; ovary superior, style exerted, stigma bifid. Fruit: drupe, globose to subglobose, 2–4 mm in diameter, reddish (*3*, *4*).

Plant material of interest: dried ripe fruits

General appearance

Mature fruit is round to ovoid, 2–4 mm in diameter, glandular hairy, extremely hard, reddish-brown to black, slightly rough, and usually accompanied by a short pedicel and some smaller, immature fruits in close groups of up to six. The apex has a slight depression with 4 faint grooves at right angles to one another. A tubular persistent calyx with 5 short, often indistinct, teeth covers half to three quarters of the surface. The calyx is grey-green and tomentose (*1*).

Organoleptic properties

Odour: faintly aromatic; taste: slightly aromatic and bitter (*4*, *9*, *10*).

Microscopic characteristics

Fruit: The exocarp is brown and narrow, consisting of parenchymatous cells with thin walls and partially lignified cells with many pitted thickenings on the inside. In surface view, the exocarp shows an epidermis of polygonal cells with thickened walls and some with large, conspicuous, simple pits; among the cells are short-stalked glandular trichomes with unicellular or multicellular heads and some short covering trichomes. In cross-section, the fruit shows small epicarp cells covered with a thick cuticle. The mesocarp consists of several layers of isodiametric parenchyma cells with slightly thickened and pitted cell walls; occasionally these cells have brownish granular contents. The walls of the outer mesocarp cells are brown whereas those of the inner cells lack colour. The inner mesocarp consists of finely pitted sclerenchymatous cells, some with moderately thickened walls, others consisting of isodiametric stone cells with a small lumen. In the outer part, very small brown-coloured vascular bundles are arranged in a circle. Towards the endocarp the cells become smaller and their cell walls thicker; the innermost cell layers consist of small sclereids with a small branched lumen. The seeds are small, having large

cotyledons surrounded by thin-walled, large parenchymatous cells that have ribbed thickenings. The nutritive tissue and the cells of the germ contain aleurone grains and oil globules. Calyx: composed of outer epidermis of small, isodiametric polygonal cells, densely covered by short, bent or undulate, unicellular or bicellular covering trichomes of fairly uniform length; inner epidermal cells a little larger, walls slightly wavy, some thickened; trichomes absent (*1*).

Powdered plant material

Greyish to dark brown, with a musty, slightly aromatic odour and unpleasant, bitter taste, reminiscent of sage; abundant, more or less isodiametric stone cells with walls of varying thickness and degree of pitting; ovoid lignified cells with thin bands of reticulate thickening; fragments of calyx with closely-spaced, short covering and glandular trichomes on the outer side and birefractive elongated sclereids on the inner side; epicarp cells with large pits in the outer wall; thin-walled parenchymatous cells and globules of fixed oil; small glandular trichomes (*1*).

General identity tests

Macroscopic and microscopic examinations (*4*, *9*, *11*), thin-layer chromatography for the presence of agnuside and aucubin (*1*), and high-performance liquid chromatography for the presence of the marker compounds, casticin and agnuside (*1*, *12*) and for the biologically active diterpenes vitexilactone, rotundifuran and 6β,7β-diacetoxy-13-hydroxy-labda-8,14-diene (*12*).

Purity tests

Microbiological

Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*13*).

Foreign organic matter

Not more than 2.0% (*1*).

Total ash

Not more than 8.0% (*1*).

Acid-insoluble ash

Not more than 2.0% (*1*).

Water-soluble extractive

Not less than 8.0% (*9*).

Loss on drying
Not more than 10.0% (*1*).

Pesticide residues
The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*13*). For other pesticides, see the *European pharmacopoeia* (*13*) and the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*14*) and pesticide residues (*15*).

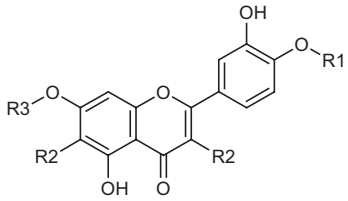
Heavy metals
For maximum limits and analysis of heavy metals, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*14*).

Radioactive residues
Where applicable, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*14*).

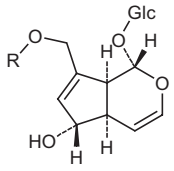
Chemical assays
Contains not less than 0.05% agnuside and 0.08% casticin calculated on the basis of dried drug by high-performance liquid chromatography (*1*).

Major chemical constituents
Up to 2.0% essential oil with bornyl acetate, 1,8-cineol, limonene, α-pinene and β-pinene being primary constituents. Flavonoids, iridoids and diterpenes represent major groups of secondary constituents found in the fruit (*4*, *5*). Casticin, in concentrations up to 0.2% (*12*) is considered the major flavonoid, with chrysosplenetin, chrysosplenol D, cynaroside, 5-hydroxy-3,4′,6,7-tetramethoxyflavone, 6-hydroxykaempferol, isorhamnetin, luteolin and luteolin 6-C-glycoside (isoorientin) derivatives being other compounds of this class. Diterpene constituents include vitexilactone (0.001–0.004%), 6β,7β-diacetoxy-13-hydroxylabda-8,14-diene, rotundifuran, and vitexlactam A (*3*, *5*, *16–18*). The structures of representative flavonoids, iridoids and diterpenes are presented below.

Medicinal uses
Uses supported by clinical data
Orally for the symptomatic treatment of gynaecological disorders including corpus luteum insufficiency and hyperprolactinaemia (*19*), premenstrual syndrome (*20–25*), menstrual irregularities (*26*, *27*), cyclic mastalgia (*28*, *29*) and also to treat hormonally-induced acne (*30*, *31*).

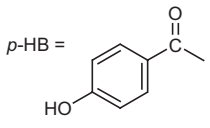


	R1	R2	R3
Casticin	CH_3	OCH_3	CH_3
Chrysosplenol D	H	OCH_3	CH_3
Cynaroside	H	H	Glc

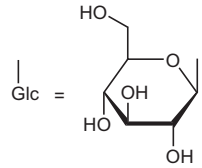


Aucubin R = H

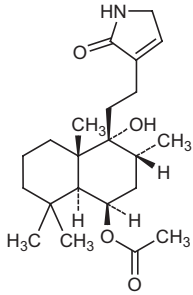
Agnuside R = *p*-HB



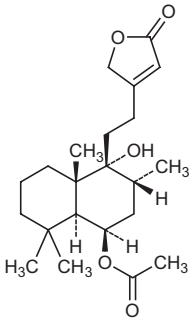
p-hydroxybenzoyl



β-D-glucopyranosyl



Vitexlactam A



Vitexilactone

Uses described in pharmacopoeias and well established documents

Orally for the treatment of endometrial hyperplasia and secondary amenorrhoea (*32*); endocrine-dependent dermatoses (dermatitis symmetrica dysmenorrhoica (Matzenauer-Polland syndrome)) acne vulgaris, eczema, acne rosacea), hypermenorrhoea (*33*), infertility due to hyperprolactinaemia and luteal phase defect (*34*). Used to treat fibroid cysts and infertility, to stop miscarriages due to progesterone insufficiency, to help expel the placenta after birth (*35*) and also as a digestive aid, sedative, anti-infective and for the treatment of hot flushes (*36*).

Uses described in traditional medicine

Used as an anaphrodisiac, calefacient, contraceptive, emmenagogue, sedative and as a tonic (*5*).

Pharmacology

Experimental pharmacology

Receptor binding

Numerous mechanisms have been proposed for the many activities of the crude drug. Extracts of the fruit have been shown to act as dopamine agonists in vitro and in vivo. The binding of an 80% ethanol extract of the fruit and various fractions of the extract to the dopamine D_2 and other receptors was evaluated both by radioligand binding studies and by super-

fusion experiments (*35*). The extract bound to the dopamine D_2 and opioid (μ and κ subtype) receptors with a range of median inhibitory concentrations between 40 and 70 μg/ml. Binding was not observed for the histamine H_1 and benzodiazepine receptor or the serotonin transporter. Two diterpenes isolated from the hexane fraction of the extract, rotundifuran and 6β,7β-diacetoxy-13-hydroxy-labda-8,14-diene, exhibited inhibitory actions on dopamine D_2 receptor binding with a median inhibitory concentration of 45 and 79 μg/ml, respectively (*16*, *37*). While lipophilic fractions of the extract bound to the μ- and κ-opioid receptors, binding to δ-opioid receptors was inhibited mainly by an aqueous fraction of the extract. In superfusion experiments, the aqueous fraction of a methanol extract inhibited the release of acetylcholine in a concentration-dependent manner. In addition, the D_2 receptor antagonist, piperone, antagonized the effect of the extract suggesting a dopaminergic action mediated by D_2 receptor activation. A labdane diterpene, α-acetoxy-13-hydroxylabdadiene, isolated from a fruit extract, was found to displace ^{125}I-sulpiride from recombinant human D_2 receptor binding sites in a dose-dependent manner (*38*). This group also demonstrated that rotundifuran, at a concentration of 100 μM, significantly inhibited the secretion of prolactin from cultured rat pituitary cells ($p < 0.05$). In addition, rotundifuran inhibited forskolin-induced prolactin and cyclic adenosine monophosphate secretion in rat pituitary cells, when added to the medium at a concentration range of 10–100 μM (*38*). Bicyclic clerodane diterpenes have also been isolated from extracts of the fruit and were found to have a 10-fold higher activity than rotundifuran for inhibiting synthesis of cyclic adenosine monophosphate and release of prolactin in prolactin secreting cells of the rat pituitary by binding directly to the D_2 receptors (*39*).

In membrane preparations from rat corpus striatum, a lyophilized 60% ethanol extract of the fruit at a concentration of 0.5 mg/ml displaced ^{125}I-sulpiride from dopamine D_2 receptor binding sites in a dose-dependent manner (*40*). An extract of the fruit as well as the synthetic dopamine agonist (lisuride) significantly inhibited basal and thyroid releasing hormone-stimulated secretion of prolactin by rat pituitary cells in vitro (*41*).

A reduction in the concentrations of endogenous opioids during the late luteal phase has also been proposed as one of the mechanisms which may induce the symptoms of premenstrual syndrome, such as mood swings, headaches and water retention (*39*). A number of fruit extracts and chromatographic fractions have been tested in vitro for their ability to displace receptor binding ligands to the μ-, κ-, and δ-opioid receptors (*37*, *42*). The extract and butanol, chloroform and hexane fractions bound to

the μ- and κ-receptors, while the aqueous extract was more active in the δ-opioid receptor. No binding in the orphan opioid receptor was noted.

Rat brain striatal tissue was preincubated with ^{3}H-choline. Treatment of the preincubated tissue with a fruit extract inhibited electrically stimulated release of ^{3}H-acetylcholine with a median inhibitory concentration of 30 μg/ml (*37*). The inhibitory effect was reduced by co-incubation of the tissues with spiroperidol. Atropine partially reduced the inhibitory effects of the fruit extract suggesting that the extract may also work on the cholinergic receptors (*37*).

Several extracts of chaste berry have been shown to bind to the estrogen receptor and have weak estrogenic effects, suggesting that chaste berry may also affect the estrogen/progesterone balance (*43–45*). A fruit extract dose-dependently bound to both estrogen receptor isotypes, but binding appeared to be more selective for estrogen receptor β than estrogen receptor α (*45*). The extract also dose-dependently inhibited the secretion of progesterone from human granuloma cells (*44*), an effect that is mediated by estrogen receptor β, as it can be blocked by tamoxifen. Furthermore one in vivo study has shown that treatment of ovariectomized rats with an undefined extract of the fruit (dose not stated) increased uterine growth, and the expression of uterine c-myc mRNA levels and liver ceruloplasm mRNA levels, indicating an estrogenic effect (*43*).

A methanol extract of the crude drug bound to both estrogen receptor α and estrogen receptor β, and induced the expression of estrogen-dependent genes, progesterone receptor, and pS2 (presenelin-2) in Ishikawa cells (an estrogen-dependent endometrial adenocarcinoma cell line) (*45*). Significant binding affinity for both estrogen receptor α and estrogen receptor β, with a median inhibitory concentration of 46.3 μg/ml and 64.0 μg/ml, respectively, and the affinity for estrogen receptor α and estrogen receptor β was not significantly different (*45*). In Ishikawa cells, the extract exhibited weak estrogenic activity, as indicated by up-regulation of the progesterone receptor mRNA; however alkaline phosphatase activity was not changed. In S30 breast cancer cells, the presenelin-2 gene was up-regulated in the presence of 20.0 μg/ml of the same extract. Based on bioassay-guided isolation, the "estrogenic" component from the fruit extract was identified as linoleic acid, which also bound to estrogen receptor α and estrogen receptor β (*46*). Like the extract, linoleic acid also induced expression of the progesterone receptor mRNA in Ishikawa cells, at a concentration of 1 μg/ml, indicating that binding produced a biological estrogenic effect in vitro. In addition, low concentrations of the extract or linoleic acid (10 μg/ml) up-regulate the expression of estrogen receptor β mRNA in the estrogen receptor+

hormone-dependent T47D:A18 cell line, a further indication of estrogenic activity (*46*).

Effect on prolactin secretion

An ethanol extract of the fruit (1:10 with ethanol, 62%), in a range of concentrations from 0.41 to 3.3 mg/ml, significantly inhibited basal and thyroid stimulating hormone-stimulated prolactin secretion from rat primary pituitary cell cultures in vitro ($p < 0.05$) (*41*, *47*). At a concentration of 3.3 mg/ml the inhibition was 80% for basal secretion and 65% for stimulated secretion. These results were confirmed in another study demonstrating significant inhibition of prolactin release from rat pituitary cells by the extract at concentrations of 0.5 mg/ml for basal secretion and 0.125 mg/ml for stimulated secretions (*41*). Furthermore, inhibition of prolactin secretion from rat pituitary cells was also observed after treatment with an extract of the fruit at concentrations of 460 µg/ml ($p < 0.0003$) for basal secretion and 115 µg/ml for stimulated secretion ($p < 0.01$) (*41*). The inhibitory effect of a fruit extract on prolactin secretion was investigated in male rats (*48*). Intravenous administration of a 53% ethanol fruit extract containing 20 mg/ml of water-soluble constituents significantly inhibited stress-induced prolactin secretion as compared with the baseline ($p < 0.05$) (*48*).

Toxicology

The median lethal dose of an ethanol extract of the fruit after a single intragastric or intraperitoneal injection was greater than 2.0 g/kg body weight (bw) in rats and mice, and no deaths were reported (*4*).

In a 28-day subacute toxicity study the no-observed-effect level was 50.0 mg/kg bw; chronic administration over 26 weeks resulted in a no-observed-effect level of 40.0 mg/kg bw (*4*). No genotoxic effects were observed when the same extract was tested in the thymidine kinase mutation assay in mammalian cell lines, the unscheduled DNA repair assay in rat hepatocytes or in the micronucleus assay of murine bone marrow cells (*4*).

Clinical pharmacology

Approximately 32 clinical trials have assessed the safety and efficacy of various fruit extracts or tinctures (53–70% ethanol) for the treatment of acne, corpus luteum insufficiency, cyclic breast pain, hyperprolactinaemia, menopausal symptoms, increasing lactation, premenstrual syndrome, uterine bleeding disorders and miscellaneous menstrual irregularities (*47*). A review of all of the clinical data is beyond the scope of this monograph; for the complete details of all trials please refer to the cited references (*4*, *47*). Most of the studies were open, uncontrolled studies investigating the effects of the extracts on menstrual cycle abnormalities or premenstrual

syndrome. One double-blind placebo-controlled study investigated a fruit extract in treatment of luteal phase defects due to hyperprolactinaemia (*19*). Two other double-blind placebo-controlled studies investigated fruit extracts in treatment of premenstrual syndrome (*24*, *49*).

Abnormal menstrual cycles and infertility

Since 1954 at least 17 studies have assessed the effects of extracts of the fruit on a variety of menstrual cycle disorders including amenorrhoea, oligomenorrhoea, polymenorrhoea, corpus luteum insufficiency and infertility (*4*). Two double-blind placebo-controlled clinical trials and several observational studies have investigated the effect of various extracts of the fruit on corpus luteal phase dysfunction and infertility (*19*, *34*). The products tested were all ethanol extracts (53–70% ethanol), and the doses used in these investigations were: 20 drops twice daily; 15 drops three times daily; 30 drops twice daily; or one to two tablets or capsules daily.

A randomized, double-blind, placebo-controlled trial involving 52 women with luteal phase defects due to latent hyperprolactinaemia assessed the efficacy of a dried fruit extract (*19*). The aim of the study was to find out whether elevated pituitary prolactin levels could be reduced and if deficits in luteal phase length and luteal phase progesterone synthesis could be normalized. Blood for hormone analysis was taken on days 5–8 and day 20 of the menstrual cycle, before and after 3 months of therapy. Latent hyperprolactinaemia was analysed by monitoring the prolactin release 15 and 30 min after intravenous administration of 200 µg of thyroid hormone. Thirty-seven cases (placebo: $n = 20$; treatment: $n = 17$) were included in the final statistical analysis. After 3 months of treatment with the extract at a dose of 20 mg per day, prolactin release was reduced; a significant increase in the length of the luteal phase (10.5 days; $p < 0.05$) was observed. Deficits in luteal progesterone synthesis were eliminated. These changes only occurred in women in the treatment group, no change was observed in the placebo group. All other hormonal parameters remained unchanged, except for 17-β-estradiol, which increased during the luteal phase in women in the treatment group. The overall length of the menstrual cycle did not change, suggesting that there was a corresponding shortening of the follicular phase. Two women in the group given the extract had become pregnant by the end of the study. No side-effects were reported.

The second randomized, double-blind, placebo-controlled study assessed the efficacy of a 53% ethanol extract of the crude drug in 96 infertile women (*34*). The outcome criteria included pregnancy or menstrual bleeding in women with secondary amenorrhoea or improved luteal hormone concentrations. The women were administered 30 drops twice daily for 3 months. Sixty-six women completed the study, but no statisti-

cally significant results were found (p = 0.069). In the women with amenorrhoea or luteal phase dysfunction, pregnancy resulted twice as often in women in the treatment group (15%) as in those in the placebo group (7%); however no statistical analysis was reported.

In open (uncontrolled) trials involving 48 women who were infertile due to luteal-phase dysfunction, the efficacy of a fruit extract for the normalization of progesterone concentrations was determined (*50*). The inclusion criteria were normal prolactin levels (below 20 ng/ml), normal results in the prolactin and thyroid-stimulating hormone stimulation tests and an abnormally low serum progesterone level (below 12.0 ng/ml) on the 20th day of the cycle. Treatment consisted of a fruit extract, 40 drops daily, without any other medication for 3 months. Forty-five women completed the studies (3 were excluded because of concurrent hormone use). The outcome of therapy was assessed by the normalization of the mid-luteal progesterone level and by correction (lengthening) of any pre-existing shortening of the phases of the cycle. Treatment was deemed successful in 39 of the 45 patients. Seven women became pregnant; serum progesterone was restored to normal in 25 patients (> 12 ng/ml) and in seven women there was a trend towards normalization of progesterone levels. However, no statistical analysis was performed.

Two larger post-marketing trials, involving 479 women, assessed the safety and efficacy of fruit extracts for the treatment of oligomenorrhoea or polymenorrhoea (*50*). The women were treated with 30 drops of the extract twice daily and the outcome measured was the bleeding-free interval. An increase in the bleeding-free interval was observed after 35 days in 187/287 women receiving treatment for oligomenorrhoea and after 26 days in 139/192 women receiving treatment for polymenorrhoea.

Acne treatment

Two uncontrolled clinical studies and one observational report have assessed the effects of extracts of the fruit on acne due to hormone imbalance (*30*, *31*, *33*). In one open study, 118 people with acne were treated with a fruit extract (20 drops twice daily for 4–6 weeks, then 15 drops twice daily for 1–2 years) and the results were compared with those of conventional treatments for acne (*31*). Patients treated with the fruit extract reported a quicker healing rate after 6 weeks and after 3 months of therapy, 70% of patients treated with the fruit extract had complete healing.

Cyclic breast pain (mastalgia)

Breast pain (mastalgia) is a common complaint usually classified as cyclical (associated with the menstrual cycle) or non-cyclical (not associated with the menstrual cycle). Mild premenstrual breast discomfort, lasting

for 1–4 days prior to menstruation that resolves upon the initiation of menstruation, is considered to be within normal physiology. Non-cyclic breast pain lasting for five or more days should be brought to the attention of a health care provider. Several open (uncontrolled) trials (*28*, *51–56*) and three randomized controlled clinical trials (*28*, *29*, *56–58*) have assessed the safety and efficacy of extracts of the fruit for the treatment of cyclic mastalgia.

A randomized, double-blind, placebo-controlled clinical trial involving 104 women with cyclic breast pain (at least 3 cycles) assessed the effects of a preparation of the fruit (tincture 1:5 equivalent to 2 g of the fruit in 53% ethanol) for the treatment of cyclic breast pain (*58*). The patients were treated with either placebo, tincture (30 drops twice daily), or tablets (one tablet twice daily) for three cycles. Patients assessed the intensity of breast pain once per cycle using a visual analogue scale and also recorded the presence of menstrual bleeding and the intensity of pain in a diary. Prolactin levels were also measured during the premenstrual week of cycles one and three. At the end of the third cycle of treatment, a significant reduction in breast pain was observed in the treated patients as compared with those who received placebo (tincture, $p = 0.006$; tablets, $p = 0.0076$). Neither the tablets nor the tincture of crude drug had any effect on concentrations of progesterone, follicle stimulating hormone or luteinizing hormone. While the basal prolactin levels decreased in both treatment groups, this was not statistically significant when compared with placebo (*58*).

A second randomized, placebo-controlled, double-blind study with a similar design compared the tincture (30 drops = 1.8 ml, twice daily for 3 cycles) with placebo for the treatment of 97 women ($n = 48$ in the treatment group; 49 in the placebo group) who had had breast pain at least 5 days prior to menses in the last cycle before the study (*57*). A visual analogue scale was used for assessment of the efficacy. Intensity of breast pain diminished more quickly in the group that received the tincture. The study design and duration were similar to that of Wuttke et al. (*57*, *58*). The results of this study showed a decrease in the visual analogue scale scores of women in both the treatment and the placebo groups. However, compared with women in the placebo group, those in the treatment group had significantly lower visual analogue scale values at the end of each cycle ($p = 0.018$, 0.006 and 0.064 for cycles 1, 2 and 3, respectively).

In a randomized, placebo-controlled trial the effects of a Vitex agnus-castus solution and a placebo (double-blind) were compared with that of gestagen (lynestrenol) in 160 women with mastalgia (*59*). A complete remission, or improvement of symptoms, was reported in 82.1%, 74.5%, and 36.8% of the patients in the gestagen, chaste tree, and placebo groups,

respectively. The difference in effect between treatment and placebo was significant ($p < 0.01$). No significant differences were found between the two treatments (*59*).

Numerous open studies have assessed the effect of a solution of *Vitex agnus-castus* (VAC solution) for the treatment of over 1700 women with mastalgia (*28*, *29*, *51*, *52*, *54–56*). All of these studies assessed the efficacy of one product, VAC solution, at a dose of 45–75 drops per day for 1–6 menstrual cycles. Two studies compared VAC treatment with lynestrenol (5 mg daily on days 12–24 of each cycle). Elimination of symptoms was observed in 46–81.5% of treated women; improvement of symptoms in 12–39.6% and no effect in 6.5–29%. Reported side-effects included circulatory disturbances, acne and weight gain.

Premenstrual syndrome

Premenstrual syndrome refers to the regular occurrence of affective symptoms such as depressive moods, irritability, anxiety, confusion and social withdrawal, as well as somatic symptoms including breast tenderness or heaviness and breast pain (mastalgia), abdominal bloating, cravings, fatigue and headache.

Twelve clinical trials have assessed the efficacy of extracts of the fruit for the symptomatic treatment of premenstrual syndrome (*22–24*, *26*, *27 49*, *58–63*). Of these studies, only three were randomized controlled trials and two were double-blind (*22*, *49*, *63*). A positive placebo effect was ruled out by one randomized placebo-controlled study carried out in compliance with good clinical practice (*63*). In this study, patients ($n = 86$) with premenstrual syndrome were given either a chaste tree fruit extract (60% ethanol), in the form of a product called "Z 440", one 20-mg tablet daily or a placebo ($n = 84$) during three consecutive menstrual cycles. Diagnosis was made according to the Diagnostic and Statistical Manual for Mental Disorders. The main efficacy variable was change from baseline to end-point (end of the third cycle) in the patient's self-assessment of six premenstrual syndrome symptoms (irritability, mood alteration, anger, headache, breast fullness, and other symptoms including bloating). A secondary efficacy variable was change in Clinical Global Impressions score for the factors: severity of condition, global improvement, and risk–benefit. Mean improvement in patient's self-assessment was significantly greater in the women in the treatment group than in women who received the placebo ($p < 0.001$). Clinical Global Impressions scores for each of the three factors also revealed significant superiority of the treatment relative to placebo ($p < 0.001$). Responder rates (> 50% reduction in symptoms) were 52% and 24% for treatment and placebo, respectively. Adverse events reported in the active treatment arm ($n = 4$) included acne, multiple

abscesses, inter-menstrual bleeding and urticaria; in the placebo arm (n = 3) the adverse events were acne, early menstrual period and gastric upset.

A randomized, double-blind, placebo-controlled trial involving 217 women with self-diagnosed premenstrual syndrome according to a modified version of the Menstrual Distress Questionnaire, a rating scale covering most of the important symptoms, assessed the efficacy of the fruit for the management of symptoms of premenstrual syndrome (*49*). Subjects were treated with either a powder of the dried fruit (300-mg tablets; two tablets three times daily; n = 105) or a soy-based placebo (n = 112) for a period of 3 months, after which they all completed the Menstrual Distress Questionnaire again. Other than a statistically significant difference in effect between the active powder and the soy-based placebo for the symptom "feel jittery and restless" (p = 0.05), no other statistically significant results were reported. Unfortunately, soy was a poor choice for use as a placebo, as it is not considered to be biologically inert.

A multi-centre, randomized, double-blind, controlled clinical trial compared the activity of a dried ethanol extract of the fruit with that of pyridoxine (vitamin B6) in the treatment of women with premenstrual syndrome (*22*). The intent-to-treat population included 127 women: 61 of whom were given one capsule of extract plus one placebo capsule daily for three cycles, while 66 were given one capsule of placebo twice daily on days 1–15 of their cycle, followed by one capsule (100 mg) of pyridoxine twice daily on days 16–35. Therapeutic response was assessed using the Premenstrual Tension Syndrome scale, the Clinical Global Impressions scale, and by recording six characteristic symptoms of premenstrual syndrome (breast tenderness, oedema, inner tension, headache, constipation and depression). Therapeutic efficacy was assessed by both patients and physicians at the end of the trial. Initial mean scores on the Premenstrual Tension Syndrome scale were higher in the group treated with the chaste tree extract (15.2) than in those treated with pyridoxine (11.9). By the end of therapy, the mean absolute change in Premenstrual Tension Syndrome score in each group was 5.1, representing a reduction of 10.1 and 6.8, respectively, for the chaste tree and pyridoxine-treated groups (p < 0.038, both groups, 95% confidence interval –6.4261 to –0.1670). Therefore no difference was evident between the two treatment groups. The Clinical Global Impressions scale showed that 77.1% of the women who received chaste berry and 60.6% of those treated with pyridoxine showed improvement. Adverse events were rare, but included gastrointestinal complaints, skin reactions and transient headache.

Six post-marketing studies assessed the safety and efficacy of various extracts of the fruit in 8391 female patients with menstrual abnormalities

or symptoms of premenstrual syndrome (*23*, *26*, *27*, *58*, *60*, *62*). Three open (uncontrolled) trials (*24*, *26*, *59*) also investigated the effect of various fruit extracts on menstrual abnormalities. The dose ranged from 40–42 drops or 1 capsule daily, for 1 day to 9 years and the outcomes measured included the physician's and patient's self-assessments. Elimination of symptoms was observed in 29–42% of patients; improvements in symptoms were observed in 51–59% of patients and symptoms were unchanged in 1–10% of patients. Adverse events were reported in 1–5% of patients and were generally not reported to be serious. The limitations of these studies include the lack of a control group and the fact that most did not distinguish between premenstrual syndrome and the other menstrual disorders.

An open (uncontrolled) clinical trial involving 50 women (43 of whom completed the study) with late luteal phase dysphoric disorder (Diagnostic and Statistical Manual for Mental Disorders) assessed the effect of an ethanol fruit extract on the management of premenstrual syndrome (*59*). Thirteen of the subjects were concurrently taking oral contraceptives. After 2 months of baseline observation, one tablet of the extract was administered daily for three cycles, followed by a post-treatment phase lasting three cycles. Treatment effectiveness was evaluated using both the Menstrual Distress Questionnaire and the visual analogue scale. The Menstrual Distress Questionnaire was filled out by patients at the end of the first cycle and again during cycles 3 and 6. The visual analogue scale was completed twice per cycle, once in the late luteal phase when symptoms peaked and once after menstruation during the follicular phase. By the end of the third cycle, the Menstrual Distress Questionnaire scores were reduced by 42.5% ($p < 0.001$), with a 50% reduction in the score in 20/43 patients. By the end of the post-treatment period, the scores remained approximately 20% below baseline ($p < 0.001$). The main improvements following treatment were reported for symptoms of breast tenderness, behavioural changes, negative feelings and oedema. The average late-luteal phase visual analogue scale score was reduced by 47.2% during the 3-month treatment phase ($p < 0.01$), and remained at 21.7% below baseline ($p < 0.001$) during the post-treatment phase. By contrast, the follicular phase score did not significantly change. The number of days with premenstrual syndrome symptoms was slightly reduced from 7.5 to 6 days, and the concomitant use of oral contraceptives had no significant effect on any of the parameters investigated. Twenty patients (47%) reported 37 adverse events during the treatment and post-treatment periods (*59*).

An open (uncontrolled) study involving 36 women with premenstrual syndrome assessed the effect of a 58% ethanol extract of the fruit for the

management of premenstrual syndrome symptoms (*24*). The women were treated with 40 drops of the extract daily over three cycles and the outcomes measured were a reduction in physical and psychological symptoms such as headache, swollen breasts, breast tenderness, bloating, fatigue and psychological changes such as increased appetite, sugar craving, nervousness and restlessness, anxiety, irritability, lack of concentration, depression, crying spells, mood changes and aggressiveness. The duration of the luteal phase was also determined. After 3 months of treatment, 69% of women had a reduction in physical symptoms and 80% showed a reduction in psychological symptoms ($p < 0.05$). The duration of the luteal phase was lengthened from 5.4 to 11.4 days. A randomized open (uncontrolled) trial assessed a tincture of the fruit (10 g tincture containing 2 g fruit in 53% ethanol) for the treatment of premenstrual syndrome. Women were treated with 30 drops twice daily in combination with vitamin E (400 mg daily). Treatment significantly reduced the symptoms of irritability or anxiety ($p = 0.028$), breast tenderness ($p = 0.0001$) and mastalgia ($p = 0.015$) (*61*).

A randomized single-blind study compared the efficacy of fluoxetine, a selective serotonin reuptake inhibitor with that of the crude drug (*64*). Forty-one patients with premenstrual dysphoric disorder according to the Diagnostic and Statistical Manual of Mental Disorders were randomly allocated to the group receiving fluoxetine or that receiving the extract for 2 months. The outcomes measured included the Penn daily symptom report, the Hamilton depression rating scale, and the clinical global impression severity of illness and improvement scales. After 2 months, 68.4% of patients had responded to fluoxetine and 57.9% to the crude drug extract. There was no statistically significant difference between the groups in the rate of responders. However, fluoxetine was more effective in alleviating the psychological symptoms, while the extract reduced the physical symptoms (*64*).

Effects on lactation

Only one randomized, double-blind controlled trial examined the effect of the fruit in lactating women (*65*). Women were treated with the fruit extract (15 drops three times daily) or vitamin B1 (no dose stated) or assigned to the control group (details not stated). Lactation in all groups increased up to day 10 postpartum; from days 10–20 a decrease in lactation was observed in women in the control and vitamin B1-treated groups. Lactation in women in the group treated with the fruit extract increased or was maintained up to day 20. Lactating women with poor milk production treated with a fruit extract were able to effectively increase production. No statistical analyses were performed.

Adverse reactions

Adverse reactions have been reported in some clinical trials. A review of 30 studies involving 11 506 subjects reported a total of 246 adverse events, thus representing an adverse reaction rate of approximately 2% (*4*). The major reactions reported included acne, changes to the menstrual cycle, dizziness, gastrointestinal distress, increased menstrual flow, nausea, skin reactions, urticaria and weight gain (*4*). Minor adverse events include fatigue, hair loss, increased intraocular pressure, palpitations, polyurea, sweating and vaginitis (*4*, *57*).

Contraindications

Fructus Agni Casti should not be used during pregnancy (*35*).

Warnings

No information available.

Precautions

General

Patients reporting a feeling of tension and swelling of the breasts or menstrual disturbances should consult a health care provider for a medical diagnosis (*66*).

Drug interactions

Although no interactions have been documented, the reported dopaminergic effect may reduce the efficacy of dopamine-receptor antagonists (*3*). Furthermore, due to its potential hormonal effects, Fructus Agni Casti may interfere with the effectiveness of oral contraceptives and hormone replacement therapy (*67*).

Carcinogenesis, mutagenesis, impairment of fertility

Intragastric administration of an ethanol fruit extract to male and female rats at doses up to 80 times the recommended human daily dose had no effect on fertility, mating behaviour, pregnancy or lactation (*3*). No pathological changes were observed in any of the offspring of treated animals when compared with those animals treated with vehicle control (*3*).

Pregnancy: non-teratogenic effects

See Contraindications.

Pregnancy: teratogenic effects

Intragastric administration of an ethanol fruit extract to rats and rabbits at doses up to 100 and 74 times higher than the human daily dose, respec-

tively, was not teratogenic and did not affect maternal health as compared with controls (*3*).

Nursing mothers

One study in rats assessed the effect of a fruit extract administered orally to lactating dams on their offspring (*68*). A decrease in milk consumption in the offspring was observed and a high rate of mortality resulted compared with untreated animals. Normal milk consumption patterns were resumed in the offspring when the dams were no longer given the extract (*68*). No further data are available; therefore the use of the crude drug by nursing mothers is not recommended.

Paediatric use

No safety data are available, therefore the use of the crude drug in children under the age of 12 years is not recommended.

Other precautions

Estrogen-dependent breast cancer patients should use Fructus Agni Casti preparations with caution, as weak estrogenic effects have been reported in vitro (*45*, *46*).

Dosage forms

Crude drug, extracts, fluidextracts, tinctures and infusions. The dried berries should be stored in airtight non-plastic containers and protected from light, heat, moisture and insect infestation (*4*).

Posology

(Unless otherwise indicated)

Dry native extract: 8.3–12.5:1 (w/w), approximately 1.0% casticin: 1 tablet containing 2.6–4.2 mg native extract, swallowed whole with some liquid each morning (*4*).

Dry native extract: 9.58–11.5:1 (w/w): 1 tablet containing 3.5–4.2 mg native extract each morning with some liquid (*22*).

Dry native extract: 6.0–12.0:1 (w/w), approximately 0.6% casticin. For premenstrual syndrome: 1 tablet containing 20 mg native extract daily with water (*63*).

Fluidextract: 1:1 (g/ml), 70% alcohol (v/v): 0.5–1.0 ml (*9*).

Tincture: ethanol 58% (100 g of aqueous-alcoholic solution contains 9 g of 1:5 tincture): 40 drops, once daily with some liquid each morning (*4*).

Tincture: ethanol 53% (10 g of the solution contains 2 g crude drug mother tincture): 30 drops twice daily (*25*, *28*, *34*).

Tablet: containing 162 mg of crude drug mother tincture (1:10 with 62% ethanol), twice daily (*57*).

Hydroalcoholic extracts (50–70% v/v): corresponding to 30–40 mg dried fruit (*4*, *69*).

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Cortex Berberidis

Definition
Cortex Berberidis consists of the dried stem bark of *Berberis vulgaris* L. (Berberidaceae) (*1*, *2*).

Synonyms
No information was found.

Selected vernacular names
Agracejo, aigret, aigribouet, almindelig berberis, ameer Paris, anbarbaris, barberry, berberi, Berberitze, berbero, berberry, berbis, cigret, cigretier, common barberry, crespino, crespino commune, epine-vinette, espino cambrón, European barberry, Gemeiner Sauerdorn, harilik kukerpuu, jaundice-berry, khichirma, kirsing, libdane, oad alreeah, piprage, pipperidge, Rocky Mountain grape, salq, Sauerdorn, skerpa, snowberry, surtorn, tarab, trailing mahonia, uqdah, vincllier, venittier, zerk, zereshk (*1*, *3–5*).

Geographical distribution
Found in Europe and North Asia, and naturalized in North America (*2*, *5*).

Description
An erect, deciduous, heavily branched thorny shrub, up to 2 metres high. The thorny branches are angular, deeply grooved, initially brownish-yellow, later becoming grey-white. The thorns are 1–2 cm long and protrude horizontally. The leaves are compound, obovate to elliptoid 2–4 cm long, and bear leaflets with dentate margins and spine. The flowers are 5–7 cm long in yellow, dense, hanging clusters. Sepals (6) are yellow and petals (6) are orange at the base. Ovary is superior with a flat stigma. The edible fruit is a bright, scarlet, oblong-cylindrical berry, 10–13 mm long and 6 mm thick, containing 2 seeds. The exocarp is membranous-coriaceous (*5*, *6*).

Plant material of interest: dried stem bark

General appearance

Crude drug may be small, flat and irregular shaped, up to 2.0 mm thick, or curved pieces up to 0.5 mm thick; outer surface dark yellowish-grey with shallow, longitudinal furrows, or thicker pieces with deeper cracks and fissures. Occasionally, black apothecia of lichens are present. Inner surface dark yellow to brown, distinctly longitudinally striated and glistening, frequently with patches of paler yellow wood attached. Fracture in the outer part is short, and readily separates, but fracture in the inner part is fibrous (*1*, *5*).

Organoleptic properties

Odour: faint aromatic; taste: bitter and imparts a yellow colour to the saliva (*1*).

Microscopic characteristics

Outer rhytidome consists of successive areas of thin-walled, lignified cork cells alternating with dark yellowish-brown areas of dead cortex and secondary phloem. The secondary phloem contains tangential bands of fibres, usually one or two cells wide, alternating with wider bands of sieve tissue and separated by medullary rays, 2–4 cells wide. The phloem fibres are small, yellow, thick walled and lignified with very numerous, conspicuous pits. Phloem consists of narrow sieve tubes and small-celled parenchyma cells. Many of the medullary ray cells contain one or occasionally two large prism crystals of calcium oxalate per cell. Other medullary ray cells contain starch granules and some medullary ray cells, especially those adjacent to the phloem fibres, will develop into stone cells with moderately thickened walls (*1*).

Powdered plant material

Yellowish-brown, containing fragments of thin-walled, lignified, polygonal cork cells; abundant short, yellow fibres occurring singly or in small groups with thick, lignified walls and very numerous pits; thin-walled sieve tubes and associated parenchyma; prism crystals of calcium oxalate in medullary ray cells and scattered, individual crystals, occasionally twinned; small, simple starch granules, rounded to ovoid; groups of rectangular stone cells with moderately thickened walls and numerous pits; abundant groups of yellowish-brown crushed parenchyma; occasional lignified fibres and vessels from the adherent xylem (*1*).

General identity tests

Macroscopic and microscopic examinations as well as thin-layer chromatography (*1*).

Purity tests

Microbiology

Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*7*).

Chemical

To be established according to national requirements.

Foreign organic matter

Not more than 2% (*1*).

Total ash

Not more than 10% (*1*).

Acid-insoluble ash

Not more than 2.5% (*1*).

Water-soluble extractive

Not less than 12% (*1*).

Alcohol-soluble extractive

To be established in accordance with national requirements.

Loss on drying

To be established in accordance with national requirements.

Pesticide residues

The recommended maximum sum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*8*). For other pesticides, see the *European pharmacopoeia* (*8*) and the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*7*) and pesticide residues (*9*).

Heavy metals

For maximum limits and analysis of heavy metals, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*7*).

Radioactive residues

Where applicable, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*7*).

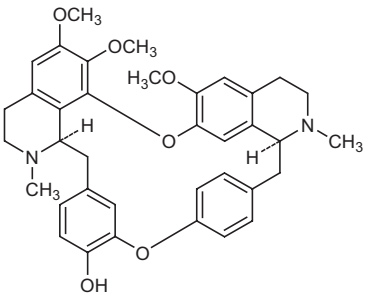
Chemical assays

To be established according to national requirements. Since the major constituents are berberine and related alkaloids, the high-performance liquid chromatography method of analysis established for berberine in Rhizoma Hydrastis (*10*) may be employed for standardization.

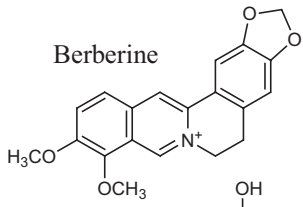
Major chemical constituents

The major constituents are isoquinoline alkaloids, principally berberine, occurring at a concentration up to 4.5%. Other major alkaloids include oxyacanthine (0.04%), berbamine (0.6–1.2%), columbamine (0.3%), jatrorrhizine (0.4%) and magnoflorine (2.1%) (*11–13*). Structures of berbamine, berberine, columbamine, jatrorrhizine and magnoflorine are presented below.

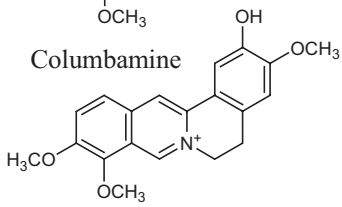
Berbamine



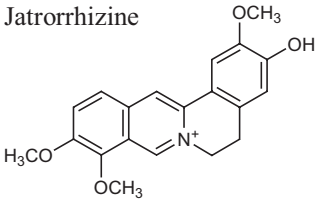
Berberine

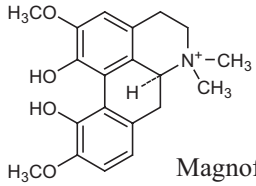


Columbamine



Jatrorrhizine





Magnoflorine

Medicinal uses

Uses supported by clinical data

None.

Uses described in pharmacopoeias and well established documents

Used orally for the treatment of digestive complaints, such as dyspepsia, diarrhoea, gastritis, feelings of distention and flatulence (*1*).

Although no clinical trials evaluating Cortex Berberidis have been conducted, the most relevant active principle is known to be berberine. Berberine has been shown to be effective for the treatment of bacterially-induced diarrhoea (*14–20*), ocular trachoma (*21*) and cutaneous leishmaniasis (*22–25*).

Uses described in traditional medicine

Used orally for the treatment of cystitis, dysmenorrhoea, eczema, fever, haemorrhoids, inflammation, menorrhagia, nasal congestion, rheumatism, tinnitus and vaginitis (*4*, *26*, *27*). Also used as a cholagogue, diuretic, emmenagogue, haemostat, laxative and a tonic (*4*).

Pharmacology

Since no clinical studies have directly evaluated Cortex Berberidis for any therapeutic effect and there have been very few pharmacological studies on extracts of the bark, this section focuses on the pharmacology and clinical studies of the major alkaloid constituents, particularly berberine. The correlation of these data with the activity of the crude drug or extracts thereof requires further investigation.

Experimental pharmacology

Antidiarrhoeal activity

In vitro, *Vibrio cholerae* grows in a medium containing berberine, but fails to produce diarrhoea-inducing toxins (*28*). It has been hypothesized that the antidiarrhoeal activity of berberine is due to localized effects on the intestinal tract and not to bactericidal activity. The suggested mechanism by which berberine exerts this action is through the activation of α_2-adrenoceptors and by reducing cyclic adenosine monophosphate production through the inhibition of the activity of adenylate cyclase (*29*), which in turn decreases intestinal motility (*30*). Berberine inhibits in vivo and in vitro intestinal secretions induced by cholera toxin (*31–34*).

Berberine, at a concentration of 10.0 mg/loop, reduced intestinal secretion stimulated by the heat-labile toxin of *Escherichia coli* in the ligated rabbit intestine loop model by 70% in situ, and inhibited the secretory response of the heat stable toxin of *E. coli* in mice (*35*, *36*). Berberine, at concentrations up to 500 µM, stimulated ion transport responses in human colonic mucosa that were non-specific for calcium ions or cyclic ad-

enosine monophosphate-mediated signals. In cultured intestinal epithelial monolayers, berberine inhibits calcium and cyclic adenosine monophosphate-mediated responses, indicating that the drug exerts a direct antisecretory effect on the epithelial cells through the inhibition of mucosal chloride secretion (*37*).

Anti-inflammatory activity

Treatment of mice with berberine, at a dose of 10.0 mg/kg body weight (bw) for 3–12 days, inhibited the development of adjuvant-induced arthritis (*38*). Berberine, administered to mice at a dose of 10.0 mg/kg bw, inhibited serotonin-induced hind paw oedema (*26*). Berberine also possessed dose-dependent antinociceptive activity, which was assessed by the inhibition of 1,4-benzoquinone-induced writhing movements as well as antipyretic activity in animals with increased rectal temperature (*26*). Intragastric administration of berberine at a dose of 7.5 or 15.0 mg/kg bw to rats with trinitrobenzene sulfonic acid-induced colitis reduced histological lesions, morphological damage and myeloperoxidase activity after 1 week of treatment (*39*).

In in vitro studies, berberine also inhibited the production of the inflammatory cytokine interleukin-8 in rectal mucosal cells at a concentration of 10^{-5} M (*39*). Treatment of a human oesophageal squamous cell carcinoma cell line, YES-2, with berberine (8.0–32.0 µM) for 24 hours reduced the expression of messenger ribonucleic acid for the inflammatory cytokine interleukin-6 (*40*).

The anti-inflammatory properties of an ethanol extract of the bark, three alkaloid fractions, the major alkaloid berberine, and oxyacanthine isolated from the crude drug were assessed after intragastric administration to mice at various doses (*41*). The outcome assessed was a reduction in the acute inflammatory response in the carrageenan- and zymosan-induced hind paw oedema model. The results indicated that ethanol extract had the greatest anti-inflammatory effects. The ability of the ethanol extract to alter in vivo and in vitro complement activity was also determined in mice. The ethanol extract also reduced chronic inflammation in this model of adjuvant arthritis. The protoberberine fractions Bv2, Bv3 and berberine suppressed a delayed-type hypersensitivity reaction. Fraction Bv1 and berberine diminished antibody response in mice (*41*).

Antiplatelet effects

Berberine significantly inhibited rabbit platelet aggregation induced by adenosine diphosphate, arachidonic acid, collagen or calcium ionophore A23187 ($p < 0.01$) (*42*). In vitro, berberine significantly inhibited synthesis of thromboxane A_2 in rabbit platelets induced by adenosine diphos-

phate, arachidonic acid or collagen, in which collagen-induced thromboxane A_2 synthesis was also inhibited (*42*). In vivo, the plasma prostacyclin level was reduced by 34.6% during a 30-min period after intravenous administration of 50.0 mg/kg bw of berberine to rabbits. These results suggest that berberine inhibits arachidonic acid metabolism in rabbit platelets and endothelial cells at two or more sites: cyclooxygenase in the arachidonic acid cascade and possibly the enzyme(s) for arachidonic acid liberation from membrane phospholipids (*42*).

Antimicrobial activity

A methanol extract of the bark and berberine inhibited the growth of *Helicobacter pylori* in vitro. Berberine inhibited the growth of 15 strains of *H. pylori,* with a minimum inhibitory concentration of 12.5 µg/ml (range 0.78–25.0 µg/ml) (*43*). The antibacterial activity of berberine against *Staphylococcus aureus* at a sub-inhibitory dose was potentiated by the flavones chrysosplenol-D and chrysoplenetin, from *Artemisia annua*. The potentiation is due to the inhibition of an *S. aureus* multidrug resistance pump (*44*). Berberine had a minimum inhibitory concentration of 25–50 µg/ml against *S. aureus* and *Mycobacterium smegmatis* (*45*, *46*). The antimicrobial activity of berberine was evaluated against 17 microorganisms including two Gram-negative bacteria, *Pseudomonas aeruginosa* and *Escherichia coli* (both resistant and sensitive), two Gram-positive bacteria, *Bacillus subtilis* and *S. aureus*, *Zoogloea ramigera*, six filamentous fungi, *Penicillium chrysogenum*, *Aspergillus niger*, *Aureobasidium pullulans* (black and white strain), *Trichoderma viride* (original green strain and brown mutant), *Fusarium nivale*, *Microsporum gypseum* and two yeasts, *Candida albicans* and *Saccharomyces cerevisiae* (*47*).

The results demonstrated that the minimum inhibitory concentration, minimum microbicidal concentration and minimum microbistatic concentrations varied considerably depending on the microorganism tested. In general, the sensitivity of the microorganisms to berberine in decreasing order was as follows: *S. aureus* > *P. aeruginosa* S (sensitive) > *E. coli* (sensitive) > *P.* aeruginosa R (resistant) > *E. coli* (resistant) > *B. subtilis* > *Z. ramigera* > *C. albicans* > S. *cerevisiae* > *A. pullulans* > *T. viride* (brown) > *M. gypseum* > *A. niger* > *F. nivale* > *P. chrysogenum* > *T. viride* (green).

Berberine weakly inhibited the growth of *Bacillus subtilis* and *Salmonella enteritidis* in vitro at a concentration of 1 mg/ml and 0.5 mg/ml, respectively (*48*). Berberine also weakly inhibited the growth of *Clostridium perfringens* in vitro (150 µg/ml), and, at a concentration of 1.0 mg/ml, weakly inhibited the growth of *Entamoeba histolytica*, *Giardia lamblia*

and *Trichomonas vaginalis* and induced morphological changes in the parasites (*49*).

Cardiovascular effects

Berberine has been reported to have protective effects against cardiac arrhythmias and severe congestive heart failure (*50–52*). This physiological effect is due to the ability of berberine to prolong action potential duration and inhibit the inward rectifier potassium current and outward delayed rectifier potassium current (50–100 μM). Berberine has also been shown to inhibit the human ether-a-go-go-related channel expression in *Xenopus* oocytes (median effective dose was 95 μM). In rats, Langendorff perfusion of an isolated heart was performed using verapamil to bring about acute heart failure. Treatment of the heart with berberine (10 μmol/l) before the use of verapamil significantly reduced the degree of heart failure as compared with the control group ($p < 0.001$) (*52*).

Effects on smooth muscle

Berberine, at a concentration of 1 μM, relaxed norepinephrine-precontracted isolated rat aorta strips (*53*). Berberine induced relaxation in isolated precontracted rat mesenteric arteries at a concentration of 10^{-5} M (*54*, *55*). At a concentration of 0.1–100 μM, berberine suppressed basal tone and induced a concentration-dependent relaxation of phenylephrine-precontracted rabbit corpus cavernosum in vitro (*56*). Intracavernous injection of berberine to anaesthetized rabbits at a dose of 5.0 mg/kg increased intracavernosal pressure from 12.7 to 63.4 mmHg, and the duration of tumescence ranged from 11.5 to 43.7 minutes (*56*). Berberine inhibited norepinephrine and phenylephrine-induced contractions in prostate strips isolated from rabbit (*57*).

The effects and mechanism of berberine on the intracellular free calcium concentration in the smooth muscle cells of guinea-pig colon were assessed in vitro using bi-wavelength spectrophotometry in suspensions of these cells. In the resting state, berberine had no significant effects on free calcium concentration, but inhibited the increase in free calcium levels induced by 60 mm potassium chloride in a concentration-dependent manner. The median inhibitory concentration was 34.09 μM. At concentrations of 30 and 100 μM, berberine inhibited the elevation of free calcium evoked by 10 μM of acetylcholine in the presence or absence of extracellular calcium. These data suggest that berberine inhibits the influx of extracellular calcium and calcium release from intracellular stores in the smooth muscle cells of colon, indicating that berberine may be a calcium channel blocker (*58*).

Immunological effects

Berberine inhibited the proliferation of mouse spleen cells induced by T-dependent mitogens (concanavalin A) and phytohaemagglutinin. Spleen cells were obtained from mice treated with berberine at a dose of 10.0 mg/kg bw for 3 days (*38*).

Intragastric administration of an extract of the cortex to rats for 6 weeks increased production of antigen-specific immunoglobulin M (*59*). Intraperitoneal administration of berberine to mice, at a daily dose of 10.0 mg/kg bw for 3 days before the induction of tubulointerstitial nephritis, significantly (p = 0.001) reduced pathological injury and improved renal function, as well as decreasing the number of CD3+, CD4+ and CD8+ T-lymphocytes as compared with controls (*60*).

Miscellaneous activity

Intraperitoneal administration of 150.0 mg/kg bw of cyclophosphamide caused serious haemorrhagic cystitis in rats after 12 hr, including bladder oedema, haemorrhage and dramatic elevation of nitric oxide metabolites in urine and in plasma. Rats were pretreated with one or two doses of berberine at 50.0, 100.0 or 200.0 mg/kg bw intraperitoneally then challenged with cyclophosphamide (150 mg/kg, intraperitoneally). The results indicated that pretreatment of rats with berberine reduced cyclophosphamide-induced cystitis in a dose-dependent manner. Furthermore, two doses of berberine showed greater protection against cyclophosphamide urotoxicity than a single dose. In addition, a single dose of 200.0 mg/kg bw berberine, or two doses of 100.0 and two doses of 200.0 mg/kg bw berberine could completely block cyclophosphamide-induced bladder oedema and haemorrhage, as well as the increase in nitric oxide metabolites in rat urine and plasma (*61*).

Toxicology

The oral median lethal dose of berberine in mice was 329 mg/kg bw (*62*). Oral administration of berberine (2.75 g) to dogs produced severe gastrointestinal irritation, profuse watery diarrhoea, salivation, muscular tremors and paralysis. Respiration was not affected. Postmortem analysis of the intestines found them to be contracted, inflamed, and empty or containing mucous and watery fluid. An oral dose of 25.0 mg/kg bw of berberine sulfate induced depression lasting for 6 to 8 hours; 50 mg/kg bw caused salivation and sporadic emesis. A dose of 100.0 mg/kg bw induced persistent emesis and death of all animals 8–10 days later (*62*). Pretreatment of rodents with a single oral dose of berberine (4.0 mg/kg bw) induced prolongation of the pentobarbital (60 mg/kg bw, intraperitoneally)-induced sleeping time as well as increased strychnine (0.3 mg/kg bw, intraperiton-

eally)-induced toxicity, suggestive of an inhibitory effect on microsomal drug metabolizing enzymes, cytochrome P450 isozymes (*63*).

Uterine stimulant effects

A hot aqueous extract of the bark (alkaloid free) stimulated contractions in isolated guinea-pig uteri at a concentration of 1:200 (1 ml extract in 200 ml bath medium) (*64*, *65*). A 70% ethanol extract of the crude drug inhibited spontaneous, oxytocin- or serotonin-induced contractions in isolated rat uteri, with a median inhibitory concentration range of 10.0–19.9 µg/ml (*66*).

Clinical pharmacology

Berberine has demonstrated efficacy for the treatment of secretory diarrhoea (*14–20*). However, the quality of the trials was limited by a lack of positive controls. Few studies have compared the efficacy of berberine with that of tetracycline for the treatment of fluid loss caused by diarrhoea in patients with cholera or in non-cholera diarrhoea (*14*, *15*, *17*, *62*).

In a randomized, double-blind, placebo-controlled clinical trial involving 400 patients with acute watery diarrhoea, the antisecretory and vibriostatic effects of berberine and tetracycline were evaluated (*14*). In this study of 185 patients with cholera, oral administration of tetracycline or tetracycline and berberine, at a dose of 100.0 mg orally four times daily, reduced the stool volume and frequency, as well as the duration of diarrhoea. In the berberine-treated group, together with a reduction in stool volume, a 77% reduction in the concentration of cyclic adenosine monophosphate in the stool was observed. Neither berberine nor tetracycline exhibited any benefit over placebo in patients with non-cholera diarrhoea of unspecified etiologies (*14*).

A randomized comparison-controlled trial involving 165 patients assessed the antisecretory activity of berberine sulfate for enterotoxigenic *Escherichia coli*-induced diarrhoea. Patients received either 400.0 mg of berberine as a single oral dose or 1200 mg of berberine sulfate (400.0 mg every 8 hours) for the treatment of cholera (*17*). In patients with *Escherichia coli*-induced diarrhoea who received a single oral dose of berberine, the mean stool volumes were significantly ($p < 0.05$) reduced over those of controls during three consecutive 8-hour periods after treatment. At 24 hours after treatment, more of the patients who were treated with berberine and had *Escherichia coli*-induced diarrhoea, had less diarrhoea than controls (42% versus 20%, $p < 0.05$). Patients with diarrhoea due to *Vibrio chlolerae* who received 400.0 mg of berberine had a reduction in stool volume, but those treated with 1200 mg of berberine plus tetracycline did not. No adverse effects were observed in the patients receiving berberine. The results of this study indicated that berberine was an effective and safe

antisecretory drug for treatment of *Escherichia coli*-induced diarrhoea, but had only a modest antisecretory effect in cholera patients, in whom the activity of tetracycline alone was superior (*17*).

A clinical study was conducted in primary-school children with ocular trachoma. Ninety-six children with this disease were selected for a blinded study. Patients were advised to instil the drops containing 0.2% berberine into the eye three times daily and to apply an ointment containing 0.2% berberine at bedtime to both eyes for a period of 3 months. The comparison drugs tested were berberine drops plus 0.5% neomycin ointment at bedtime; 20% sodium sulfacetamide drops with 6% sodium sulfacetamide ointment at bedtime, or placebo (normal saline) drops. The children treated with berberine had a response rate of 87.5%, whereas those treated with berberine and neomycin had a response rate of 58.83%. The study showed that 83.88% were clinically cured but only 50% became microbiologically negative when treated with berberine alone (*21*).

Berberine has been used therapeutically for the treatment of cutaneous leishmaniasis, commonly referred to as "oriental sore", by subcutaneous injection of berberine near the site of the lesion (*22–24*). In patients with cutaneous leishmaniasis caused by *Leishmania tropica*, injection of a preparation containing 2% berberine into lesions was an effective treatment (*22*, *23*).

Pharmacokinetics and pharmacodynamics

In animal models, berberine has poor bioavailability because it is a quaternary ammonium compound. It has been suggested that the poor bioavailability of berberine may be due to the activation of P-glycoprotein (*67*). In an investigation on enhancing its bioavailability, the effect of P-glycoprotein inhibitors cyclosporin A, verapamil and the monoclonal antibody C219 were investigated using in vivo and in vitro models of intestinal absorption to determine the role of P-glycoprotein in berberine absorption (*51*). In the rat re-circulating perfusion model, berberine absorption was improved sixfold by co-administration of P-glycoprotein inhibitors. In the rat intestinal sac model, berberine serosal-to-mucosal transport was significantly decreased by cyclosporin. In Ussing-type chambers, the rate of serosal-to-mucosal transport across rat ileum was threefold higher than in the reverse direction and was significantly decreased by cyclosporin. In Caco-2 cells, berberine absorption was significantly increased by P-glycoprotein inhibitors and by monoclonal antibody C219. P-glycoprotein appears to contribute to the poor intestinal absorption of berberine, indicating that P-glycoprotein inhibitors could be of therapeutic value by improving its bioavailability (*51*).

The pharmacokinetics of berberine were assessed by high-performance liquid chromatography coupled to microdialysis to determine the concentration of unbound berberine in rat blood, liver and bile (*68*). Microdialysis probes were simultaneously inserted into the jugular vein towards the right atrium, the median lobe of the liver and the bile duct of rats for biological fluid sampling after administration of berberine, at a dose of 10 mg/kg bw, through the femoral vein. A linear concentration–response relationship was observed over the concentration range 0.05–50.0 µg/ml. The results demonstrated that the disposition of berberine occurred in the blood, liver and bile fluid. These data indicate that berberine is metabolized in the liver and undergoes hepatobiliary excretion (*68*).

The structures of the unknown metabolites of berberine were determined after isolation of the compounds from the urine of five volunteers. Each of the volunteers was given an oral dose of berberine chloride, 0.9 g per day, for 3 days. Metabolites in urine samples were isolated and purified by polyporous resin column chromatography and identified by electrospray ionization mass spectroscopy and proton nuclear magnetic resonance spectroscopy. The three major unknown metabolites (M1, M2 and M3) were isolated and identified as jatrorrhizine-3-sulfate, demethyleneberberine-2-sulfate and thalifendine-10-sulfate (*50*).

Toxicology and overdose

Following accidental ingestion by humans of more than 500 mg of berberine, lethargy, nosebleeds, dyspnoea, and skin and eye irritation may occur. In addition, kidney irritation, nephritis and lethal poisoning have been reported. Disturbances of the gastrointestinal tract with nausea, vomiting and diarrhoea have also been observed (*69*).

Adverse reactions

No information found.

Contraindications

Cortex Berberidis is contraindicated in patients with hypersensitivity or allergy to the plant material.

Warnings

Exposure of patients to sunlight or artificial light sources emitting UVA should be avoided when topical preparations derived from the crude drug or berberine are applied (*70*).

Precautions

General

Use with caution in patients with high blood pressure, diabetes, glaucoma or a history of cardiovascular disease.

Drug interactions

Berberine is reported to upregulate the expression of the human multidrug resistance gene coding for multidrug resistance transporter (PGP-170); thus the treatment of tumours with berberine may result in reduced retention of chemotherapeutic agents such as paclitaxel (*71*, *72*). Berberine has been reported to interact with cyclosporin in renal transplant patients (*73*). Blood concentrations of cyclosporin were enhanced by 75% after co-administration of berberine hydrochloride in renal transplant patients, but this did not increase the toxicity of cyclosporin (*73*).

Drug and laboratory test interactions

None reported.

Carcinogenesis, mutagenesis, impairment of fertility

The genotoxic effects of berberine in prokaryotic cells were assessed in the SOS chromotest in *Saccharomyces cerevisiae* (*74*). No genotoxic activity with or without metabolic activation was observed, and no cytotoxic or mutagenic effects were seen under nongrowth conditions. However, in dividing cells, the alkaloid induced cytotoxic and cytostatic effects in proficient and repair-deficient *Saccharomyces cerevisiae.* In dividing cells, the induction of frameshift and mitochondrial mutations, as well as crossing over, showed that the compound is not a potent mutagen (*74*).

Pregnancy: teratogenic effects

No information was found.

Pregnancy: non-teratogenic effects

Due to a lack of safety data, the use of the crude drug during pregnancy is not recommended.

Nursing mothers

Due to a lack of safety data, the use of the crude drug during breastfeeding is not recommended.

Paediatric use

Due to a lack of safety data, the use of the crude drug in children under the age of 12 years is not recommended.

Dosage forms

Crude drug and dried extracts, fluidextracts, and tinctures (*1*, *75*). Store in a tightly sealed container away from heat and light.

Posology

(Unless otherwise indicated)
Daily dose: crude drug 0.5–1.0 g three times daily, or by decoction; liquid extract 1:1 in 60% ethanol, 0.3–1.0 ml three times daily; tincture 1:10 60% ethanol, 2–4 ml three times daily (*1*).

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Gummi Boswellii

Definition
Gummi Boswellii consists of the dried gum resin of *Boswellia serrata* Roxb. ex Colebr. (Burseraceae) (*1*).

Synonyms
Boswellia glabra Roxb., *B. thurifera* (Colebr.) Roxb. (*2*, *3*).

Selected vernacular names
Alberodell'incenso, anduga, arbore à encerns, boswellia, boswellie-dent-elee, chilakdupa, dhupelio, dhup-gugali, dhupdo, fan hun hsiang, fan hun shu, gajabhakshya, gandhabiroz, gobahr shalla, gugal, guggul, guggula, husn-e-lubban, Indian frankincense tree, Indian olibanum, Indischer-weihrauch, kapitthaprani, kondagugi tamu, kondor, koonkanadhoopam, kundre, kundrikam, kundur, kundur luban, kunduru, kunthreekan, kunturukkam, labana, loban, loban zakar, lobhan, luban, luban-dacar-hindi, luban dhakar, maddi, madi, madimar, pahadi, parangisambrani, parangisampirani, saladi, salai, Salaibaum, salai cha dink, salai gonad, salai guggal, salakhi, saleda, saledhi, saledo, salgai, sallaki, sambrani, samprani, sanlaki, shaledum, shallaki, susrava, tallaki, vishesha dhoop, visheshdhup, zarw (*1*, *2*, *4–7*).

Geographical distribution
Native to India (*3*, *6*, *7*).

Description
A medium to large deciduous tree, up to 18 m in height and 2.4 m in girth. Leaves imparipinnate, leaflets ovate or ovate-lanceolate, variable. Flowers small, white, in axillary racemes or panicles. Drupes 12 mm long, trigonous, scarlet when young, turn white at maturity. Bark thick and aromatic. When cut, a secretion exudes and becomes gum-like after exposure to air (*1*, *2*, *7*).

Plant material of interest: dried gum-resin

General appearance

The gum solidifies slowly with time. It is reddish brown, greenish yellow, or dull yellow to orange in colour. It occurs in small, ovoid, fragrant tears. Sometimes the tears form agglomerated masses up to 5 cm long and 2 cm thick. Fracture is brittle, fractured surface is waxy and translucent. Burns readily and emanates an agreeable characteristic balsamic resinous odour (*1*, *2*).

Organoleptic properties

Odour: aromatic, characteristically balsamiferous; taste: agreeable (*1*, *2*).

Microscopic characteristics

Debris of fibres, rectangular cork cells, very few yellowish oil globules and numerous, small or large, oval to round or rhomboidal crystalline fragments present (*2*).

Powdered plant material

Not applicable.

General identity tests

Macroscopic examinations (*1*, *2*), microchemical and fluorescence tests and thin-layer chromatography (*1*), high-performance liquid chromatography (*8*, *9*) and gas chromatography–mass spectrometry for the presence of boswellic acids (*10*), as well as gas chromatography–mass spectrometry analysis for volatile and semi-volatile terpenes (*11*).

Purity tests

Microbiological

Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*12*).

Foreign organic matter

Not more than 5% (*1*).

Total ash

Not more than 10% (*1*).

Acid-insoluble ash

Not more than 8% (*1*).

Water-soluble extractive

Not less than 28% (*1*).

Alcohol-soluble extractive
Not less than 45% (*1*).

Loss on drying
To be established in accordance with national requirements.

Pesticide residues
The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*13*). For other pesticides see the *European Pharmacopoeia* (*13*) and the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*12*) and pesticide residues (*14*).

Heavy metals
For maximum limits and analysis of heavy metals, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*12*).

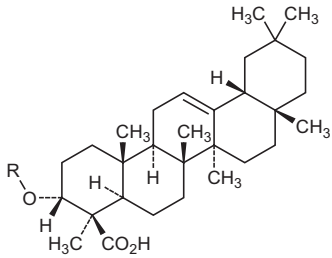
Radioactive residues
Where applicable, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*12*).

Chemical assays
To be established in accordance with national requirements.

Major chemical constituents
Contains 5–9% essential oil with major constituents being α-thujene (50–61%), sabinene (5%), α-pinene (8%) and α-phellandrene (2%). Major triterpene constituents of biological interest are members of the boswellic acids (more than 12) including 11-oxo-β-boswellic acid, 3-*O*-acetyl-11-oxo-β-boswellic acid, α-boswellic acid, β-boswellic acid, 3-*O*-acetyl-α-boswellic acid, and 3-*O*-acetyl β-boswellic acid (*3*, *15–17*). The structures of representative boswellic acids are presented below.

Medicinal uses
Uses supported by clinical data
Orally for the management of arthritis, bronchial asthma, Crohn's disease and ulcerative colitis (*18–22*).

Uses described in pharmacopoeias and well established documents
Orally for the treatment of rheumatism and arthritis (*1*, *2*, *23*).

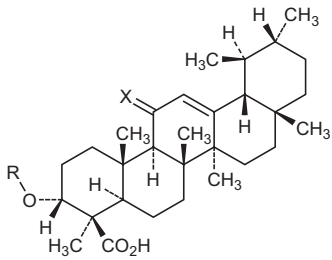


α-Boswellic acid

R = H

3-*O*-Acetyl-α-boswellic acid

R = CO-CH_3



β-Boswellic acid

R = H X = H_2

3-*O*-Acetyl-β-boswellic acid

R = CO-CH_3 X = H_2

11-Oxo-β-boswellic acid

R = H X = O

3-*O*-Acetyl-11-oxo-β-boswellic acid

R = CO-CH_3 X = O

Uses described in traditional medicine

Treatment of abdominal pain, asthma, coughs, dysentery, fever, jaundice, kidney stones, pimples, sores and stomach disorders (*5*). Also used as an antivenin and an emmenagogue (*24*, *25*).

Pharmacology

Experimental pharmacology

Analgesic activity

Intragastric administration of the gum at doses ranging from 100.0–500.0 mg/kg body weight (bw) had no analgesic effects in dogs, rabbits or rats (*26–28*). However, administration of a non-phenolic fraction of the crude drug produced analgesia in 60% of rats treated with a dose of 60.0 mg/kg bw. A dose of 150.0 mg/kg bw induced analgesia in 70% of rats (*29*). The degree of analgesia was comparable to a dose of 3–4.5 mg/kg bw of morphine. A dose of 150.0 mg/kg bw also caused a 70% reduction in spontaneous motor activity that lasted for 2 hours (*29*).

Anticomplementary activity

Boswellic acids exhibited anticomplementary activity in vitro, as assessed by the reduction of immune-induced haemolysis of antibody-coated sheep erythrocytes by pooled guinea-pig serum (*30*). The decrease in immune-induced haemolysis was due to inhibition of C3-convertase of the classical complement pathway. The threshold concentration for inhibiting C3-convertase was found to be 100.0 µg/ml (*30*).

Anti-inflammatory activity

The anti-inflammatory activity of an aqueous extract of the crude drug was assessed in vivo. The extract significantly inhibited both the maximal oedema response and the total oedema response during 6 hours of carrageenan-induced rat paw oedema (*31*). Intragastric administration of the gum or an aqueous methanol extract of the gum (9:1) to rats, at a dose of 50.0–200.0 mg/kg bw reduced carrageenan- or adjuvant-induced pedal oedema by 34–73% (*26*, *32*). An ethanol extract of the crude drug, administered at a dose of 50.0–200.0 mg/kg bw, exhibited anti-inflammatory activity in carrageenan-induced oedema in rats and mice and dextran-induced oedema in rats (*28*). The extract also had considerable anti-arthritic activity but no significant effect was observed in the cotton pellet-induced granuloma test. Treatment with the extract inhibited inflammation-induced increase in serum transaminase levels and leukocyte counts, but lacked any analgesic or antipyretic effects in rats (*28*).

The anti-inflammatory effects of the crude drug and boswellic acids were assessed in rats with adjuvant-induced arthritis. The animals were treated with 100.0 mg/kg bw of the crude drug or 200.0 mg/kg bw of boswellic acid administered by gastric lavage for 2 weeks (*33*). The activity of β-glucuronidase was used to assess lysosomal stability, which is an important factor in the arthritic syndrome. Induction of arthritis reduced lysosome stability, but treatment with either the extract or boswellic acid increased stability and had a protective effect on lysosomal integrity (*33*). Specific boswellic acids inhibit elastase in leukocytes, inhibit proliferation, induce apoptosis and inhibit topoisomerases of leukaemia and glioma cell lines (*23*).

A methanol extract of the crude drug, containing boswellic acids and their structural derivatives, was applied topically to the backs of mice to determine its anti-inflammatory effects (*23*). The treatment markedly inhibited 12-*O*-tetradecanoylphorbol-13-acetate-induced skin inflammation, epidermal proliferation, the number of epidermal cell layers and tumour promotion in 7,12-dimethylbenz[*a*]anthracene-initiated mice. Feeding 0.2% of crude drug in the diet to CF-1 mice for 10–24 weeks reduced the accumulation of parametrial fat pad weight under the abdomen, and inhibited azoxymethane-induced formation of aberrant crypt foci by 46% (*22*).

Boswellic acids (15.0 µg/ml) inhibit the biosynthesis of the pro-inflammatory leukotrienes in neutrophilic granulocytes by a non-redox, non-competitive inhibition of 5-lipoxygenase, an enzyme in the pro-inflammatory arachidonic acid cascade (*34–36*). The extract and its derivatives caused a concentration-dependent decrease in the formation of leukotriene

B4 from endogenous arachidonic acid in rat peritoneal neutrophils in vitro (*37*). Acetyl-11-keto-β-boswellic acid induced the most pronounced inhibition of 5-lipoxygenase product formation, with a mean inhibitory concentration (IC_{50}) of 1.5 µM. However, boswellic acid in concentrations of up to 400.0 µM did not inhibit cyclooxygenase or 12-lipoxygenase in isolated human platelets, or peroxidation of arachidonic acid by Fe-ascorbate. These data suggest that boswellic acids are specific, nonreducing-type inhibitors of the 5-lipoxygenase product formation either interacting directly with the 5-lipoxygenase or blocking its translocation (*37*). 3-*O*-acetyl-11-keto-β-boswellic acid is the most potent inhibitor of 5-lipoxygenase. In vitro, 3-oxo-tirucallic acid inhibited 5-lipoxygenase product synthesis in cell-free assays, with an IC_{50} of 3 µM, but in intact cells enhanced 5-lipoxygenase product formation in ionophore-challenged polymorphonuclear leukocytes and initiated calcium mobilization, MEK-1/2 phosphorylation and 5-lipoxygenase translocation at a concentration of 2.5 to 15 µM. These data indicate that the pentacyclic triterpenes that inhibit 5-lipoxygenase in cell-free systems activate 5-lipoxygenase product formation in intact cells (*38*).

Extracts of the crude drug and its constituents, the boswellic acids, activate the mitogen-activated protein kinases p42 and p38 in isolated human primed polymorphonuclear leukocytes in vitro (*39*). Activation of mitogen-activated protein kinases was rapid and transient with maximal activation after 1–2.5 min of exposure and occurred in a dose-dependent manner. The keto-boswellic acids (11-keto-β-boswellic acid and 3-*O*-acetyl-11-β-keto-boswellic acid) activated kinase at 30.0 µM, whereas other boswellic acids lacking the 11-keto group were less effective. Moreover, 11-keto-boswellic acids induced rapid and prominent mobilization of free Ca^{2+} in polymorphonuclear leukocytes. Inhibitor studies revealed that phosphatidylinositol 3-kinase is involved in boswellic acid-induced mitogen-activated protein kinase activation, whereas a minor role was apparent for protein kinase C. Mitogen-activated protein kinase activation by 3-*O*-acetyl-11-β-keto-boswellic acid was partially inhibited when Ca^{2+} was removed by chelation (*39*).

Mixed acetyl-boswellic acids significantly inhibited ionophore-stimulated release of the leukotrienes B4 and C4 from intact human polymorphonuclear leukocytes, with 50% inhibitory concentration (IC_{50}) values of 8.48 µg/ml and 8.43 µg/ml, respectively. Purified acetyl-11-keto-β-boswellic acid was about three times more potent as an inhibitor of the formation of both leukotriene B4 (IC_{50}, 2.53 µg/ml) and leukotriene C4 (IC_{50}, 2.26 µg/ml) from human polymorphonuclear leukocytes in the same assay. Daily intraperitoneal administration of an extract of mixed

acetyl-boswellic acids (20.0 mg/kg bw) significantly reduced the clinical symptoms in guinea-pigs with experimental autoimmune encephalomyelitis between days 11 and 21. However, the inflammatory infiltrates in the brain and the spinal cord were not significantly less extensive in the treated animals than in the respective control group. The multiple intraperitoneal administrations of boswellic acids did not inhibit the ionophore-challenged ex vivo release of leukotrienes B4 and C4 from polymorphonuclear leukocytes separated from the blood of guinea-pigs with experimental autoimmune encephalomyelitis or the biosynthesis of leukotrienes in vitro (*40*). Suspensions of rat peritoneal polymorphonuclear leukocytes elicited with glycogen, stimulated by calcium and ionophore to produce leukotrienes and 5-15-hydroxyeicosateraenoic acid (HETE) from endogenous arachidonic acid, were treated with various concentrations of an ethanol extract of the crude drug. A concentration-dependent inhibition of leukotriene B4 and 5-HETE production was observed. All products of 5-lipoxygenase from endogenous arachidonic acid in polymorphonuclear leukocytes were reduced (*41*).

Miscellaneous activities

Intragastric administration of a 50% ethanol extract of the crude drug, at a dose of 250.0 mg/kg bw, to rats reduced blood glucose levels (*42*). Intravenous administration to dogs of a 50% ethanol extract, at a dose of 50.0 mg/kg bw, reduced blood pressure (*42*). Intragastric administration of the crude drug at a dose of 100.0 mg/kg bw to cockerels, rabbits and rats fed a high cholesterol diet reduced serum cholesterol by 25–45% (*26*). Intragastric administration of the crude drug, at a dose of 100.0–500.0 mg/kg bw did not reduce fever in rats, rabbits or dogs (*26*). Acetyl-11-keto-beta-boswellic acid, a constituent of a herbal medicine from *Boswellia serrata* resin, attenuated experimental ileitis in animals (*43*).

Toxicology

The gestation period or parturition time in pregnant rats and the onset time of castor oil-induced diarrhoea were unaffected by the extract and no significant effect was seen on cardiovascular, respiratory and central nervous system functions. Intragastric administration of the crude drug to dogs, rabbits or rats, at a dose of 500.0 or 1000.0 mg/kg bw, did not have ulcerogenic effects. The oral and intraperitoneal median lethal doses were greater than 2.0 g/kg bw in mice and rats (*28*). Intragastric administration of the crude drug to monkeys (500.0 mg/kg bw), mice (2.0 g/kg bw) or rats (1.0 g/kg bw) for 6 months produced no observable behavioural, biochemical or histological abnormalities (*44*).

Clinical pharmacology

A double-blind pilot study, involving 37 patients with rheumatoid arthritis, assessed the effects of the crude drug on the symptoms of swelling and pain, and amount of self-medication with nonsteroidal anti-inflammatory drugs (*41*). Patients were treated with 3.6 g of the crude drug or placebo for 12 weeks. Outcome measures included Ritchie's index for swelling and pain, and the dose of nonsteroidal anti-inflammatory drugs patients felt they needed. There were no subjective, clinical or laboratory parameters showing significant changes from baseline to 12 weeks of treatment (*45*).

A randomized double-blind, placebo-controlled cross-over study was conducted to assess the efficacy, safety and tolerability of a crude *Boswellia serrata* extract in 30 patients with osteoarthritis of the knee. Fifteen subjects received the active *Boswellia serrata* extract or placebo for 8 weeks. After the first treatment, a washout period was permitted and the groups were then crossed over to receive the opposite intervention for 8 weeks. All patients receiving *Boswellia serrata* extract reported a decrease in knee pain, increased knee flexion and increased walking distance. The frequency of swelling in the knee joint was decreased, but radiology detected no change. The observed differences between drug treatment and placebo were statistically significant ($p < 0.05$). *Boswellia serrata* extract was well tolerated by the subjects except for minor gastrointestinal adverse events (*22*).

A double-blind, placebo-controlled study involving 40 patients with bronchial asthma assessed the effects of an extract of the crude drug for treatment of symptoms. The patients were treated with a preparation of gum resin of 300 mg three times daily for a period of 6 weeks. After treatment, 70% of patients showed improvement as evident from the disappearance of physical symptoms and signs such as dyspnoea, bronchial asthma and decreased number of attacks, as well as a decrease in eosinophil count and electron spin resonance. In the control group (treated with lactose, 300.0 mg three times daily, for 6 weeks), only 27% of patients in the control group showed improvement. The data show a role for the crude drug in the treatment of bronchial asthma (*20*).

A randomized, double-blind, controlled, parallel group comparison clinical trial involving 102 patients assessed the effects of the crude drug for the treatment of Crohn's disease (*18*). The positive control arm was treated with mesalazine. The primary outcome measure was the change in the Crohn's Disease Activity Index between the time of enrolment and the end of therapy. The Crohn's Disease Activity Index was reduced by 90 after treatment with the crude drug and by 53 after treatment with mesalazine; however, the difference between the two treatments was not

statistically significant. Thus, the study concluded that an extract of the crude drug was as effective as mesalazine for the treatment of Crohn's disease.

The effect of an extract of the crude drug was assessed in patients with ulcerative colitis, grade II or III (*19*). Patients were treated with the gum resin preparation (350 mg three times daily for 6 weeks), and the effects on stool properties, histolopathology and scan microscopy of rectal biopsies, blood parameters including haemogloblin, serum iron, calcium, phosphorus, proteins, total leukocytes and eosinophils were measured. Patients receiving sulfasalazine (1 g three times daily) served as controls. At the end of the treatment period, all parameters tested had improved after treatment with *Boswellia serrata* gum resin and the results were similar to those in the control group: 82% of treated patients went into remission following treatment with the gum resin as did 75% of those treated with sulfasalazine (*19*).

A controlled clinical trial assessed the efficacy of the crude drug in patients with chronic colitis characterized by vague lower abdominal pain, bleeding per rectum with diarrhoea and palpable tender descending and sigmoid colon. Thirty patients with chronic colitis, 17 men and 13 women aged between 18 and 48 years, were included in the study. Twenty of the patients were given a preparation of the gum resin of *Boswellia serrata* (300 mg three times daily for 6 weeks) and 10 were given sulfasalazine (1 g three times daily for 6 weeks) and served as controls. Of 20 patients treated with *Boswellia* gum resin, 18 showed an improvement in one or more of the parameters: stool properties; histopathology; scanning electron microscopy; and measures of haemoglobin, serum iron, calcium, phosphorus, proteins, total leukocytes and eosinophils. Of 20 patients treated with *Boswellia* gum resin, 14 went into remission whereas in the patients treated with sulfasalazine, the remission rate was 4 out of 10 (*20*).

Adverse reactions

Minor gastrointestinal side-effects have been reported in the clinical trials (*19–22*).

Contraindications

Hypersensitivity or allergy to the crude drug.

Warnings

No information was found.

Precautions

General

No information was found.

Drug interactions

None reported.

Drug and laboratory test interactions

None reported.

Carcinogenesis, mutagenesis, impairment of fertility

No information was found.

Pregnancy: teratogenic effects

No information was found.

Pregnancy: non-teratogenic effects

Due to a lack of safety data, the use of the crude drug during pregnancy is not recommended.

Nursing mothers

Due to a lack of safety data, the use of the crude drug during breastfeeding is not recommended.

Paediatric use

Due to a lack of safety data, the use of the crude drug in children under the age of 12 years is not recommended.

Dosage forms

Crude drug, extracts.

Storage

Store in a cool dry place away from heat or light.

Posology

(Unless otherwise indicated)
Crude drug: 1–3 g daily (frequency not specified) (*1*).
Extracts: 300–350 mg three times daily (*19–21*).

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Semen Cardamomi

Definition

Semen Cardamomi consists of the dried ripe seed of *Elettaria cardamomum* (L.) Maton (Zingiberaceae) recently removed from the capsule (*1–9*).

Note: In a number of formularies and standards, the drug is official as Fructus Cardamomi, but the part employed is the seed, with the fruit capsule removed prior to use (*5–10*).

Synonyms

Amomum repens Sonn., *A. cardamomum* Lour. (*11*), *Alpinia cardamomum* Roxb. (*12*).

Selected vernacular names

Alaicha, bach dau khau, bastard cardamom, cardamomier, Cardamompflanze, cardamomo minore, cardamomo, cardamon, cardamone, cardamone petite, Cardamonen, chinne elakulu, chittelam, chhoti lachi, chota elaich, chotaa leicaha, choti ilayachi, Echte Kardamon, elã, elachi, elakay, elakkki, elam, elayachi, elchi, Elettarie, ensal, ga qolia, garidimong, gujurati, haal boa, habbahan, habbahal, hab el haal, heel khurd, hel, hhabb el hal, hiliki, illlachi, kakilahekurd, kakula saghira, kapoelaga, kardoem, Kleine Kardamomen, krako, küçük kakule, lahanveldoda, lesser cardamom, luuk en, malabar cardamon, Malabar Cardamonen, pelaga, qaquilah saghirah, ronde kardemon, sa nhon, sanna elakulu, sanna yalakii, sarooplaachi, shozuku, siruelam, so du gu, sodugo, syouzuku, truti, velchi, velloda (*2–6*, *12*, *13*).

Geographical distribution

Native to India and introduced to Sri Lanka (*14*, *15*).

Description

A perennial zingiberaceous herb attaining a height of 2–4 m, with lanceolate leaves borne on long sheathing stems. Flowers numerous, borne on

horizontal racemes that arise from the rhizome and run horizontally along the ground. The fruit is inferior, ovoid or oblong, nearly ellipsoidal, capsule plump or slightly shrunken, the seeds of which are covered by an aril (*14*).

Plant material of interest: dried seed (in dried or nearly dried fruit)

General appearance

Usually in agglutinated groups of 2–7 seeds. Each irregularly angular, 3- to 4-sided, oblong, ovoid; 2–4 mm long, up to 3 mm broad; pale orange to dark reddish-brown; usually enveloped by a thin colourless membranous aril externally, transversely wrinkled but not minutely pitted; hilum depressed; raphe indicated by a channel extending on one side from base to apex; hard; internally, whitish, showing a thin dark testa, a whitish starchy perisperm grooved on one side, and in the centre a small yellowish translucent endosperm, surrounding a paler minute embryo (*1–3*, *5–7*).

Fruit: Inferior, ovoid or oblong, nearly ellipsoidal, capsule plump or slightly shrunken; 8–20 mm, but most fruits are 10–15 mm long, 5–10 mm in diameter; green to pale buff, sometimes yellowish-grey, mostly 3-sided; externally, smooth or longitudinally striated; base, rounded and may bear the remains of the stalk; apex, more or less blunt and sometimes crowned by a short beak formed of the remains of the floral parts; interior longitudinally divided into 3 loculi, each loculus is an adherent mass of two rows of 3–7 small seeds attached to the axile placenta (*2*, *5–7*). According to Evans (*15*), four varieties of fruits are available: "Mysore", "Malabar", "Mangalore" and "Alleppy".

Organoleptic properties

Odour: aromatic; taste: aromatic, pungent and slightly bitter (*1–3*, *5–8*).

Microscopic characteristics

Aril of several layers of elongated, more or less collapsed, flattened, thin-walled parenchyma cells containing small rounded or oval droplets of oil. Testa with an outer epidermis of long, narrow fusiform cells, 20–30 µm wide, having slightly thickened undulating walls, followed by a layer of collapsed parenchymatous cells, with brownish contents, becoming 2–3 layers in the region of the raphe, composed of large thin-walled, rectangular cells containing volatile oil; a band of 2–3 layers of parenchymatous cells and an inner epidermis of thin-walled flattened cells. Inner integument consists of 2 layers of cells, an outer layer of yellowish to reddish-brown, rectangular, strongly lignified sclereids, about 40 µm long and 20 µm wide, nearly filled with a small warty nodule of silica and an in-

ner epidermis of flattened cells. Perisperm of thin-walled cells, packed with minute starch grains, 1–6 µm in diameter, and containing 1–7 small prisms of calcium oxalate, about 10–20 µm long. Endosperm, of small thin-walled parenchymatous cells, each filled with a hyaline or granular mass of protein but no starch; embryo, of small thin-walled cells containing aleurone grains; fibrous sclerenchyma and large vessels present in pericarp (*2*, *3*).

Powdered plant material

Reddish to greyish-brown, characterized by numerous fragments of perisperm cells, each filled with starch granules and containing one or more prisms of calcium oxalate, 10–25 µm long; polyhedral masses of adherent starch granules from perisperm, individual granules, up to 4 µm in diameter; numerous fragments of yellowish to reddish-brown sclereids; occasional particles of epidermal cells, often crossed at right angles by the cells of the collapsed layer (*2*).

General identity tests

Macroscopic and microscopic examinations (*1–3*, *5–8*, *14*).

Purity tests

Microbiology

Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*16*).

Chemical

To be established in accordance with national requirements.

Foreign organic matter

Not more than 3% (*2*, *17*).

Total ash

Not more than 6% (*3*, *6–8*, *17*).

Acid-insoluble ash

Not more than 4% (*1*, *3*, *5–8*).

Water-soluble extractive

Not less than 10% (*3*).

Alcohol-soluble extractive

Not less than 2% (*3*).

Loss on drying
To be established in accordance with national requirements.

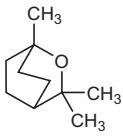
Pesticide residues
The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*18*). For other pesticides, see the *European Pharmacopoeia* (*18*) and the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*16*) and pesticide residues (*19*).

Heavy metals
For maximum limits and analysis of heavy metals, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*16*).

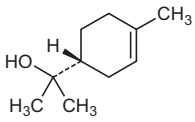
Radioactive residues
Where applicable, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*16*).

Chemical assay
Contains not less than 3.3% essential oil (v/w) (*5–8*).

Major chemical constituents
Contains 2–8% essential oil, the major constituents of which are 1,8-cineole (20–40%), (+)-α-terpinyl acetate (30–42%), α-terpineol (4–45%), limonene (6%), and smaller amounts of linalool and linalool acetate, among others (*13*, *20–22*). The structures of 1,8-cineole, (+)-α-terpinyl acetate and α-terpineol are presented below.

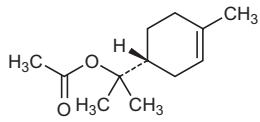


1,8-Cineole



α-Terpineol

and enantiomer



(+)-α-Terpinyl acetate

Medicinal uses
Uses supported by clinical data
None.

Uses described in pharmacopoeias and well-established documents
Orally for the treatment of dyspepsia (*23*, *24*).

Uses described in traditional medicine

Treatment of asthma, bronchitis, colic, coughs, fainting, fever, rheumatism, stomach cramps and urinary stones. Also used as an aphrodisiac, appetizer, diuretic and emmenagogue (*13*, *24*).

Pharmacology

Experimental pharmacology

Anti-inflammatory and analgesic activities

An in vivo study was performed to compare the anti-inflammatory activity of the essential oil of the seed, at doses of 175 and 280 µl/kg body weight (bw) with that of indometacin at a dose of 30.0 mg/kg bw against acute carrageenan-induced plantar oedema in rats. Intraperitoneal administration of 280 µl/kg bw of the essential oil to rats or 233 µl/kg bw to mice suppressed carrageenan-induced pedal oedema (*25*).

One study assessed the analgesic activity of the essential oil from the seed using 1,4-benzoquinone as a chemical stimulus for pain in mice. Intragastric administration of a dose of 233 µl/kg bw of the essential oil produced a 50% reduction in writhing (stretching syndrome) induced by intraperitoneal administration of a 0.02% solution of 1,4-benzoquinone (*25*). Butanol and ether extracts of the seeds had anti-inflammatory activity in vitro, as assessed in the albumin stabilizing assay (concentration not stated). The aqueous extract of the seeds, however, was not active (*26*).

Antimicrobial effects

At a concentration of 500 ppm, the essential oil from the seed weakly inhibited the growth of the fungi *Arthroderma simii*, *Chaetomium indicum*, *Microsporum canis* and *Trichophyton mentagrophytes* in vitro (*27*).

Antispasmodic activity

A 95% ethanol extract of the crude drug at a concentration of 200 µg/ml reduced histamine-induced and barium chloride-induced contractions in guinea-pig ileum in vitro (*28*). Administration of essential oil from the seed to rabbits, at a dose of 0.4 ml/kg bw, inhibited acetylcholine-induced intestinal spasms (*25*). However, an aqueous extract of the crude drug was reported to stimulate the rectus abdominus muscle of the frog and rat jejunum in vitro at a concentration of 10% of the bath media (*29*).

Smooth-muscle relaxant activity

The essential oil from the seed relaxed isolated guinea-pig ileum and trachea in vitro with a median effective dose of 15 mg and 27 mg/l, respectively. An aqueous ethanol seed extract (10 mg/ml in bath medium) relaxed guinea-pig ileum in vitro (*30*).

Antiulcer activity
Intragastric administration of dried aqueous or methanol extracts of the seeds to mice, at a dose of 53.0–126.9 mg/kg bw, reduced the secretion of gastric juice (*31*). A methanol extract of the dried seeds (5:1) at a concentration of 100 µg/ml inhibited the growth of *Helicobacter pylori* in vitro (*32*).

Choleretic activity
Intraduodenal administration of a dried acetone extract of the dried seed to rats at a dose of 50.0–500.0 mg/kg bw, had significant choleretic effects ($p < 0.01$–0.05) (*33*).

Central nervous system depressant activity
Intraperitoneal administration of the essential oil from the seed, at a dose of 1.6 ml/kg bw, induced drowsiness and a staggering gait in mice (*25*).

Dermatological effects
Application of the essential oil from the seed at a concentration of 1.0% to rabbit skin enhanced the dermal penetration of piroxicam, indometacin and diclofenac sodium (*34*). Application of the essential oil from the seed, at a dose of 1.0 ml/per square inch enhanced the dermal penetration of indometacin in rats, rabbits and humans (*34*, *35*). External pre-application of the essential oil from the seed, at a concentration of 5%, to rabbit skin increased the bioavailability of piroxicam gel (*34*).

Toxicity
Intragastric or subcutaneous administration of an aqueous ethanol (1:1) extract of the seed, at a dose of 10 g/kg bw had no observed toxicity in mice (*36*).

Miscellaneous activities
An aqueous ethanol (1:1) extract of the seed, at a concentration of 10.0 mg/ml had histaminergic activity in guinea-pig ileum in vitro (*30*). Aqueous extracts (10% w/v) of the crude drug were used for an ex vivo study of their effect on gastric secretion. The stomach of pentobarbitone-anaesthetized rats was perfused at 0.15 ml/min with aqueous extracts of cardamom or acetylcholine (1.0 µg/ml or 10.0 µg/ml solutions, in 40-minute time periods, twice in each experiment) followed by saline perfusions. The acid content in the samples was estimated by titration with 0.1 N sodium hydroxide with phenolphthalein as indicator. Acute gastric mucosal injury was induced by leaving aspirin 125.0 mg/kg bw in the stomach for 2 h before perfusion. The aqueous extract significantly increased gastric secretion from 0.10 to 0.28 ($p < 0.005$) (*37*).

Adverse reactions
Contact dermatitis has been reported (*38*).

Contraindications
Hypersensitivity or allergy to the crude drug.

Warnings
No information was found.

Precautions
General
Patients with gallstones should consult their health care provider before using preparations of Semen Cardamomi (*23*).

Drug interactions
None reported.

Drug and laboratory test interactions
None reported.

Carcinogenesis, mutagenesis, impairment of fertility
A hot aqueous or methanol extract of the crude drug at concentrations of 50 mg/disc was not mutagenic in the Ames test in *Salmonella typhimurium* strains TA98 and TA100 (*39*). The essential oil inhibited formation of DNA adducts with aflatoxin B1 by inhibiting activation in rat liver microsomes, thus demonstrating antimutagenic activity. The median inhibitory concentration was 10 µl/disc (*40*).

Drug interactions
No information was found.

Drug and laboratory test interactions
None reported.

Pregnancy: teratogenic effects
No information was found.

Pregnancy: non-teratogenic effects
No information was found.

Nursing mothers
No information was found.

Paediatric use
No information was found.

Dosage forms and storage
Crude drug, extracts and tinctures (*23*).

Posology
(Unless otherwise indicated)
Average daily dosage: 1.5 g of drug; equivalent preparations.
Tincture: daily dosage equivalent to 12 g (*23*).

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Fructus Chebulae

Definition

Fructus Chebulae consists of the dried fruits of *Terminalia chebula* Retz. or *T. chebula* Retz. var. *tomentella* Kurt. (Combretaceae) (*1–3*).

Selected vernacular names

Abhaya, ahlilaj kâbuli, alalekai, alayla, amagola, arabi, aralu, areyra, aridadi, badamier chebule, bal har, black myrobalan, bush kaduka, chebulic myrobalan, Chebulische Myrobalane, divya, Ga ja, Habra, hacha, halela, halela kabuli, halela zard, halileh, halileh kaboli, halilehsiyâh, halileh zard, hallilaj, harad, harar, harda, hardo, harir, haritaki, harra, harro, harroh, haser, helikha, hezi, himaja, hirda, hirdo, hireda, hlilej khel, hlijej sfer, hokikha, ihlilaj kabuli, inknut tree, jivathi, kabuli-harda, hora, kadukka, kadukkai, kale har, karaka, karakkaya, kashi, katukka, kãyasthã, kot-pung-pla, kurka, medicine terminalia, mirobalan de caboul, mirobalano, myrobalan, myrobalano nero, myrobalans, myrobaran, pathyã, pile har, pilo-harde, post-e-haleela kabli, post-e-haleela siyah, post-e-haleela zard, pulo-harda, rispiger Myrobalanenbaum, rong mao he zi, silikha, sa-mo-thai, samo-thai, shajar shiir hindi, sirri hindi, silikha, sivã, sringitiga, suddha, terminaalia, vayastha, vijayã, yellow myrobalan, yellow myrobalan plum, zama, zangli har (*2–11*).

Geographical distribution

Native to Cambodia, China, India, Lao People's Democratic Republic, Malaysia, Myanmar, Philippines, Thailand and Viet Nam and cultivated elsewhere (*4*, *9*, *10*, *12*).

Description

A tropical shade tree, usually 15–20 m high, but can be up to 30 m in height, and up to 1.3 m in girth; bark rough, scaly; shoots and young leaves usually rusty villous. Leaves simple, opposite, coriaceous, broadly ovate to ovate-elliptic, 7–15 cm in width by 8–25 cm in length, glabrescent; veins obscure above, slightly raised and usually brownish pubescent beneath;

apex acute or abruptly acuminate; base cuneate, slightly cordate or rounded; petiole 1–3 cm long, glabrous or sparsely pubescent with a pair of nodular glands near leaf base. Inflorescences axillary or terminal panicles, usually with 3–6 spikes (each 3–6 cm long); rachis pubescent; flowers 2 mm long, 3–4 mm in diameter; bracts nearly glabrous, 1.5–2.0 mm long; calyx outside glabrous, inside densely villous, calyx-segments triangular; stamens 3–4 mm long; ovary glabrous, ovoid, 1 mm long; style glabrous, 2.5–3.0 mm long; disc lobed, densely villous. Fruit a drupe, glabrous, subglobose to ellipsoid, 2.5–5.0 cm by 1.5–2.5 cm, usually smooth or frequently 5-angulate, ridged, wrinkled, turning blackish when dry. Seed: one, rough, ellipsoid, 1.0–2.0 cm by 0.2–0.7 cm, and without ridges (*9*).

Plant material of interest: dried fruits

General appearance

Oblong or ovoid, 2.5–5.0 cm in length, 1.5–2.5 cm in diameter. Externally yellowish-brown or dark brown, somewhat lustrous, marked with 5 or 6 longitudinal ribs and irregular wrinkles, base with a rounded fruit stalk scar. Texture compact. Sarcocarp 2–5 mm thick, yellowish-brown or dark yellowish-brown; kernels 1.5–2.5 cm long, 1.0–1.5 cm in diameter, pale yellow, rough and hard. One seed, narrowly fusiform, 1.0–2.0 cm by 0.2–0.7 cm; testa yellowish-brown, cotyledons 2, white, overlapping and convolute (*1*, *2*, *8*).

Organoleptic properties

Odour: slight and characteristic (*8*); taste: bitter, sour, astringent, then sweet (*1*, *8*).

Microscopic characteristics

Transverse section of the fruit shows epicarp composed of a layer of epidermal cells, the outer tangential wall and upper portion of the thick radial walls. Mesocarp, 2 or 3 layers of collenchyma followed by a broad zone of parenchyma with fibres and sclereids in groups, and vascular bundles, scattered; fibres, simple-pitted walls; porous parenchyma; sclereids, various shapes and sizes, mostly elongated; tannins and aggregate crystals of calcium oxalate in parenchyma; starch grains simple rounded or oval in shape, measuring 2–7 µm in diameter. Endocarp consists of thick-walled sclereids of various shapes and sizes, mostly elongated. Fibres, sclereids and vessels, lignified. Testa, one layer of large cubical cells, followed by a zone of reticulate parenchyma and vessels; tegmen consists of collapsed parenchyma. Cotyledon folded and containing aleurone grains, oil globules and some rosette aggregate crystals (*2*).

Powdered plant material
Brownish in colour and shows the diagnostic characteristics of the unground drug (*2*).

General identity tests
Macroscopic (*1*, *2*, *8*) and microscopic examinations (*2*), microchemical test (*8*), and thin-layer chromatography (*1*) and high-performance capillary electrophoresis for the presence of the marker tannins chebulinic and chebulagic acids (*13*).

Purity tests
Microbiological
Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*14*).

Foreign organic matter
Not more than 1% (*2*).

Total ash
Not more than 5% (*1*).

Acid-insoluble ash
Not more than 1% (*1*).

Water-soluble extractive
Not less than 30% (*1*).

Alcohol-soluble extractive
Not less than 30–40% (*2*, *8*).

Loss on drying
Not more than 14% (*8*).

Pesticide residues
The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*15*). For other pesticides, see the *European Pharmacopoeia* (*15*) and the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*14*) and pesticide residues (*16*).

Heavy metals
For maximum limits and analysis of heavy metals, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*14*).

Radioactive residues

Where applicable, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*14*).

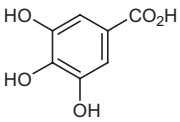
Chemical assays

To be established in accordance with national requirements.

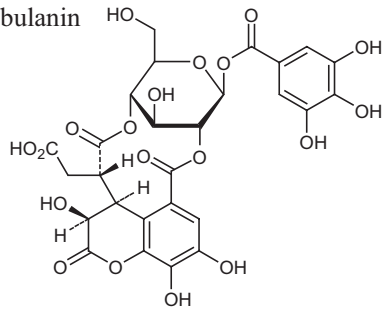
Major chemical constituents

Major constituents of the fruit are hydrolysable tannins and components thereof, including chebulagic acid, chebulinic acid, chebulanin, corilagin, gallic acid, gallic acid methyl ester, punicalagin, terchebulin and terminalic acid. Flavonols of interest include quercetin, isoquercitrin and rutin (*6*). Structures of chebulagic acid, chebulinic acid, chebulanin and gallic acid are presented below.

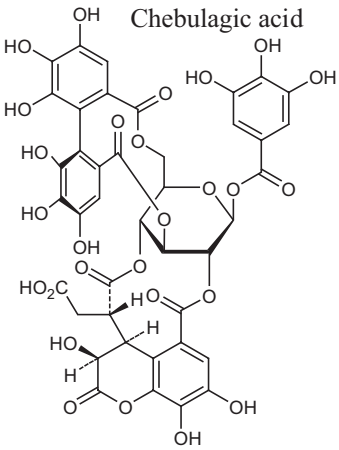
Gallic acid



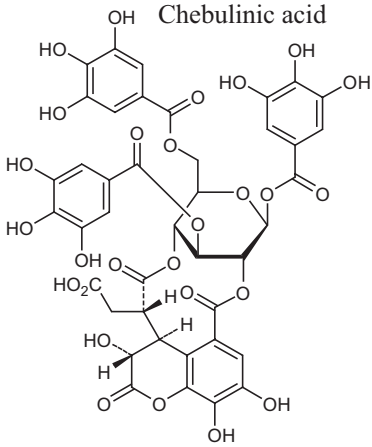
Chebulanin



Chebulagic acid



Chebulinic acid



Medicinal uses

Uses supported by clinical data

None.

Uses described in pharmacopoeias and well established documents

Used orally to treat cough with sore throat, as well as diarrhoea (*1*).

Uses described in traditional medicine

Used orally as an anthelminthic, astringent, cardiotonic, dentifrice, diuretic and laxative. Also used to treat bleeding gums, diabetes, gastrointestinal disorders, ulcers and urinary disorders (*6*).

Pharmacology

Experimental pharmacology

Antiallergic activity

An aqueous ethanol (1:1) extract of the fruit exhibited antihistamine and antispasmodic activities at a concentration of 10 mg/ml in guinea-pig ileum (*17*). The effect of an aqueous soluble fraction of a fruit extract (AF) was investigated in models of systemic and local anaphylaxis (*18*, *19*). Oral administration of AF 1 hour before injection of compound 48/80, inhibited compound 48/80-induced anaphylactic shock by 100% when AF was administered at doses of 0.01–1.0 g/kg body weight (bw). When the extract was administered 5 or 10 min after injection of compound 48/80, the mortality also decreased in a dose-dependent manner. In addition, passive cutaneous anaphylaxis was inhibited by 63.5 ± 7.8% after oral administration of the aqueous extract at a dose of 1.0 g/kg bw. In vitro, AF, in a concentration range of 0.01–1.0 mg/ml also significantly suppressed compound 48/80-induced histamine release from rat peritoneal mast cells ($p < 0.01$), and significantly increased production of tumour necrosis factor-alpha induced by anti-dinitrophenyl IgE (*19*).

Antimicrobial activity

An aqueous extract of the fruit (concentration not stated) was active against six dermatophytes, namely *Trichophyton mentagrophytes*, *T. rubrum*, *T. soudanense*, *Candida albicans*, *Torulopsis glabrata* and *C. krusei* in vitro (*20*). The in vitro antibacterial activity of an extract of the crude drug was assessed in the disc diffusion assay. The extract was active (concentration range 30–500 µg/disc) against human pathogenic Gram-positive and Gram-negative bacteria, including *Shigella dysenteriae*, *S. flexneri*, *S. boydii*, *Proteus mirabilis*, *P. vulgaris*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa* and *Salmonella* species (*21*). A 50% ethanol ex-

tract of the fruit inhibited the growth of methicillin-resistant *Staphylococcus aureus* (MRSA), with a minimum inhibitory concentration of 31.3 µg/ml (*22*).

The effect of ether, alcohol and aqueous extracts of the fruit on *Helicobacter pylori* was assessed using the agar diffusion method. An aqueous extract of the fruit inhibited the growth of the bacterium with a minimum inhibitory concentration of 125 mg/l and a minimum bactericidal concentration of 150 mg/l. Aqueous extracts, at a concentration of 1–2.5 mg/ml, also weakly inhibited urease activity of *H. pylori* (*23*).

Treatment with the powdered fruit significantly suppressed murine cytomegalovirus load in the lungs of treated mice compared with mice that received water treatment. Intragastric administration of 750 mg/kg bw per day of the dried fruit increased the body weight of infected mice and reduced the virus yield in the lungs (*24*).

The fruit showed stronger anti-herpes simplex virus type 1 activity when used in combination with acyclovir (*25*). When acyclovir and/or the extract were administered to mice by gavage at doses corresponding to those used in humans, the combinations significantly limited the development of skin lesions and/or prolonged the mean survival times of infected animals as compared with that of animals that received either acyclovir or the extract alone ($p < 0.01$ or 0.05). The combinations were not toxic to mice. The extract reduced virus yields in the brain and skin more than acyclovir alone and exhibited stronger anti-herpes simplex virus type 1 activity in the brain than in the skin, in contrast to treatment with acyclovir alone (*25*).

A hot aqueous extract of the fruit was active against anti-herpes simplex virus and was also examined for anti-cytomegalovirus activity in vitro and in vivo. The extract inhibited the replication of human cytomegalovirus and murine cytomegalovirus in vitro and inhibited plaque formation of human cytomegalovirus at a median effective concentration of 2.3 µg/ml. The anti-cytomegalovirus activities of the extract were also examined, in immunosuppressed mice. Mice were treated with various doses of cyclosporin, and immunosuppression and murine cytomegalovirus infection were monitored by measuring suppression of antibody production and virus load in the lung. The extract (15 mg/day) was administered intragastrically to mice which had been treated with 50 mg/kg bw of cyclosporin from one day before intraperitoneal infection. Concomitant administration of the extract reduced the viral load in the lung (*26*).

The effect of a methanol extract of the fruit on HIV-1 reverse transcriptase was assessed. The extracts showed significant inhibitory activity with a median inhibitory concentration (IC_{50}) ≤ 6 µg/ml (*27*).

The inhibitory activity of a hot aqueous extract of the fruit against HIV-1 protease was assessed in vitro (*28*). The extract inhibited the activity of HIV protease at a concentration of 25 µg/ml (*28*).

Antihyperlipidaemic activity

The effect of intragastric administration of an extract of the fruit (500.0 mg/kg bw for 45 days) was investigated in a model of experimental atherosclerosis in rabbits fed a cholesterol-rich diet (*29*). Atherosclerotic lesions of the aorta were examined histologically and hyperlipidaemia was assessed. Treatment of the rabbits with the extract significantly reduced cholesterol, phospholipids and triglyceride levels as compared with those in control animals ($p < 0.05$), and reduced atherosclerotic lesions.

The effect of an extract of the fruit on cholesterol-induced hypercholesterolaemia and atherosclerosis was investigated in rabbits (*30*). The control group was fed a high-cholesterol diet alone whereas the treatment group was fed both the extract and a high-cholesterol diet. Hypercholesterolaemia was significantly less ($p < 0.001$) in the treated group (166 mg/dl) than in the control group (630 mg/dl). Aortic sudanophilia was significantly less after treatment (6%), than in the control group (38%) ($p < 0.001$). The cholesterol contents of the liver and aorta were significantly less in the treatment group (46 mg/100 g and 28 mg/100 g, respectively), than in the control group (604 mg/100 g and 116 mg/100 g) (*30*).

Antioxidant activity

Various extracts (butanol, chloroform, ethyl acetate and methanol) and the isolated pure compounds: casuarinin, chebulanin, chebulinic acid and 1,6-di-*O*-galloyl-β-D-glucose from the crude drug were investigated for anti-lipid peroxidation, anti-superoxide radical formation and free radical scavenging activities in vitro. The results showed that all tested extracts and isolated pure compounds of the crude drug exhibited active to weakly active antioxidant activity at different potencies. Median inhibitory concentrations ranged from 0.005–5.39 mg/ml for the extracts and 0.031–7.27 mg/ml for the pure compounds (*31*).

The antioxidant activity of an aqueous extract of the crude drug, as estimated by thiobarbituric acid reactive substances, was tested by studying the inhibition of radiation-induced lipid peroxidation in rat liver microsomes at different doses in the range of 100–600 µg/ml. The IC_{50} in this assay was 14.5 µg/ml. The extract was also found to restore the antioxidant enzyme superoxide dismutase following radiation-induced damage. The median inhibitory activity of the extract was 11.5 µg/ml in the 1,1-diphenyl-2-picrylhydrazyl radical scavenging assay (*32*).

Cardiovascular effects
A 90% ethanol extract of the fruit increased cardiac output and had positive inotropic effects in isolated frog hearts, when added to the bath media at concentrations of 0.3 to 3.0 mg/ml (*33*).

Gastrointestinal activity
The effect of the dried powdered fruits on gastrointestinal motility in rats was assessed. The animals were divided into four groups as follows: group 1 (n = 15), normal animals; group 2 (n = 6), rats administered metoclopramide (1.35 mg/kg bw); group 3 (n = 8), rats given atropine (0.45 mg/kg bw). These agents were injected intramuscularly, 30 minutes before the experiment. Rats in group 4 (n = 8) were administered the dried fruits by gavage at a dose of 100 mg/kg/day for 15 days before the experiment. All rats were then given a test meal of methyl cellulose (1.5%) mixed with phenol red (50 mg/100 ml) orally, and gastric emptying was measured 20 minutes later. Gastric emptying of normal rats (group 1) was found to be 51.6 ± 7.79%. Treatment with metoclopramide (group 2) significantly increased the gastric emptying (76.33 ± 12.37%; $p < 0.01$) and treatment with atropine (group 3) inhibited the motility (gastric emptying 7.26 ± 19.76%; $p < 0.01$). Administration of the powdered crude drug (group 4) increased the gastric emptying (86.57 ± 6.65%; $p < 0.01$) (*34*).

Intragastric administration of the crude drug to rats, at a dose of 1.5 g/l for 15 days, reduced the number of gastric ulcerations induced by pentagastrin and carbachol (*35*).

Immunosuppressive effects
Gallic acid and chebulagic acid were isolated from a fruit extract as active chemical constituents that block cytotoxic T lymphocyte (CTL)-mediated cytotoxicity. Gallic acid and chebulagic acid weakly inhibited the killing activity of a CD8+ CTL clone with an IC_{50} of 30 µM and 50 µM, respectively. Granule exocytosis in response to anti-CD3 stimulation was also blocked by gallic acid and chebulagic acid at equivalent concentrations (*36*).

Toxicology
Dietary administration of the fruit to rats, as 25% of the diet, produced hepatic lesions which included centrilobular vein abnormalities and centrilobular sinusoidal congestion. Marked renal lesions were also observed, and included marked tubular degeneration, tubular casts and intertubular congestion. A brown pigmentation of the tail and limbs was also observed after 10 days (*37*). The median lethal dose of a 50% ethanol extract of the fruit was 175.0 mg/kg bw after intraperitoneal administration (*38*).

Clinical pharmacology
No information was found.

Adverse reactions
No information was found.

Contraindications
Hypersensitivity or allergy to the plant material.

Warnings
No information was found.

Precautions
General
No information was found.

Carcinogenesis, mutagenesis, impairment of fertility
Extracts of the fruit are not mutagenic, but have antimutagenic activities in various experimental systems. Aqueous, chloroform and acetone extracts were tested in the Ames histidine reversion assay using TA98 and TA100 tester strains of *Salmonella typhimurium* against the direct-acting mutagens, 4-nitro-*o*-phenylenediamine and sodium azide, and the indirect-acting promutagen, 2-aminofluorene, in the presence of phenobarbitone-induced rat hepatic S9 enzymes. The chloroform and acetone extracts inhibited mutagenicity induced by both direct-acting mutagens and by S9-dependent mutagens. A significant inhibition of 98.7% was observed with the acetone extract against the revertants induced by the S9-dependent mutagen, 2-aminofluorene, in a co-incubation mode of treatment (*39*).

The antimutagenic activity of a tannin fraction (TC-E) from the dried fruit pulp of the crude drug was evaluated against two direct-acting mutagens, 4-nitro-*o*-phenylenediamine and 4-nitroquinoline-*N*-oxide, and S9-dependent mutagen, 2-aminofluorene, in TA98 and TA100 strains of *Salmonella typhimurium*. The results showed that the extract (TC-E) and its fractions were antimutagenic against the S9-dependent mutagen, 2-aminofluorene. The effective concentrations ranged from 8.9–320 µg/ml (*40*).

The antimutagenicity of aqueous and chloroform extracts of the fruit were determined against two direct-acting mutagens: sodium azide in strains TA100 and TA1535, and 4-nitro-*o*-phenylenediamine in TA97a

and TA98 strains of *Salmonella typhimurium*, and the S9-dependent mutagen 2-aminofluorene in the TA97a, TA98 and TA100 strains. The aqueous extract reduced 4-nitro-*o*-phenylenediamine- as well as 2-aminofluorene-induced his^+ revertants, but had no perceptible effect against sodium azide-induced his^+ revertants in TA100 and TA1535 strains of *S. typhimurium* (*41*).

Pregnancy: non-teratogenic effects
Due to a lack of safety data, the use of the crude drug during pregnancy is not recommended.

Nursing mothers
Due to a lack of safety data, the use of the crude drug during breastfeeding is not recommended.

Paediatric use
Due to a lack of safety data, the use of the crude drug in children under the age of 12 years is not recommended.

Dosage forms
Crude drug and extracts.

Posology
(Unless otherwise indicated)
Daily dosage: 3–9 g of crude drug for decoction in divided doses (*1*). Store in an airtight container in a dry place (*1*).

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Semen Cucurbitae

Definition

Semen Cucurbitae consists of the dried seeds of *Cucurbita pepo* L. (Cucurbitaceae) or its cultivars (*1–3*).

Synonyms

Cucurbita aurantia Willd., *C. courgero Ser.*, *C. esculenta* Gray, *C. fastuosa* Salisb., *C. melopepo* L., *C. ovifera* L., *C subverrucosus* Willd., *C. verrucosus* L., *Pepo melopepo* Moench., *P. verrucosus* Moench., *P. vulgaris* Moench. (*4*, *5*).

Selected vernacular names

Abobora, bitter bottle gourd, bucka, calabaza, cubini, duraffere, dubba, dynia, étkezési tök, Garten-Kürbis, geonwomu, ghia kaddu, giramonte, giraumon, gourd, græskar, guicoy, harilik kõrvits, herkules-keule, jerimum, kadu, kadu I maghrebi, kadu I rumikao montini, kaula, kurlaru, kumra, lob-abyad, lob-kar-e-asal, malange, mandelgræskar, marrow, navadna buca, ntite, ntsuudya, pepokabocha, pompion, pompoen, pottaigummadi, pumpkin, qar, qar maghrebi, qar rumi, qara'a, safed kaddu, Schmuckkürbis, shada kumra, summer pumpkin, uritök, zapallo, zapayo, zerri at l-ger-a, zucca indi, zucchette, zucchini (*6–8*).

Geographical distribution

Native to North America and cultivated worldwide (*4*, *9*, *10*).

Description

Annual, running, monoecious herbs with dark green, non-glossy, 3–5 lobed leaves; prostrate or climbing; branched, prickly stems, up to 10 m long. The solitary flowers are large and yellow being arranged singly in the axils of leaves; the male flowers have a peduncle of 10–17 cm, a calyx with very small sepals, a campanulate deep yellow corolla (7–10 cm in diameter) gradually widening towards the top. Calyx lobes are narrow. Female flowers are similar to the male ones, but with a shorter peduncle, small stamin-

odes, and inferior ovary of various shapes. The gourd-fruit varies in size (15–40 cm in diameter) and shape in the many cultivated varieties, and the toughened, furrowed peduncle does not enlarge near it (*4*, *10*).

Plant material of interest: dried seeds

General appearance

The seeds are ovate, constricted at one end forming a short, blunt extension; flat or weakly biconvex; up to 25 mm long and 8–14 mm wide, 3–4 mm thick; on both faces, close to the edge, is an encircling ridge and groove, 1–2 mm wide, absent from projection; testa creamy-white to pale beige with a satiny sheen, smooth or with irregular wrinkles; texture brittle, somewhat papery; inner surface of seed coat fawnish-white, dull, rough or scurfy. The seed is non-endospermic. Embryo easily separated from testa, more or less entirely covered in a dark olive-green pellicle, with metallic lustre; light patches of inner seed coat may be adherent. Embryo pale greenish-yellow, oily; large, almost flat cotyledons, small conical radical at constricted end of seed; inner surfaces of cotyledons with three or five rudimentary veins, palmately arranged (*11*).

Organoleptic properties

Odour: indistinct; taste: bland, oily and slightly nut-like (*2*, *10*).

Microscopic characteristics

Epidermal cells of testa erect, prismatic, up to 200 µm long; walls thin, bearing slender vertical strips of thickening, usually sinuous in upper portion; in surface view polygonal, large with conspicuous beads; starch grains abundant, up to 5 µm, simple but frequently clumped; a band, about six cells deep, of small, thin-walled, isodiametric or small, elongated parenchymatous cells, finely reticulately thickened and strongly lignified; a few larger, irregular simple pits; a single layer of large, sub-rectangular sclereids, lumen narrow, ovoid, walls very thick and conspicuously layered, pits few and not well-defined, only middle lamella and primary wall strongly lignified; in surface view the sclereids are somewhat elongated and the anticlinal walls deeply sinuous. Internal to the sclereid band several layers of progressively larger lignified parenchymatous cells with very fine reticulate thickening; the cells, having short arm-like projections, form a spongy, lacunose tissue; areas of contact between branches of cells have quite large simple perforations. Innermost layers less well-defined, parenchymatous, largest cells internally; greenish chromoplasts present. Cotyledon cells variable, very thin-walled, containing oily globules and aleurone grains up to 4 µm in diameter (*2*).

Powdered plant material
To be established in accordance with national requirements.

General identity tests

Macroscopic and microscopic examinations (*2*, *4*, *10*), and thin-layer chromatography (*2*).

Purity tests

Microbiological
Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*11*).

Foreign organic matter
Not more than 1% (*1*).

Total ash
Not more than 7% (*2*).

Acid-insoluble ash
To be established in accordance with national requirements.

Water-soluble extractive
To be established in accordance with national requirements.

Loss on drying
Not more than 12.0% (*1*).

Pesticide residues
The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*12*). For other pesticides, see the *European pharmacopoeia* (*12*) and the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*11*) and pesticide residues (*13*).

Heavy metals
For maximum limits and analysis of heavy metals, consult the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*11*).

Radioactive residues
Where applicable, consult the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*11*).

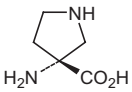
Chemical assays

To be established in accordance with national requirements.

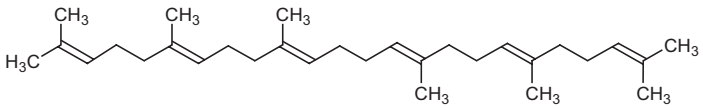
Major chemical constituents

Major constituents of the seed include a fixed oil (30–53%), phytosterols (1%) and terpenes. The main fatty acids found in the fixed oil are linoleic acid (35–68%) and oleic acid (15–48%). Squalene is a characteristic constituent and accumulates in the non-saponifiable fraction of the oil to a concentration of 39–46%. The primary phytosterols are Δ^7-sterols including spinasterol, α-spinasterol, Δ^7-avenasterol, Δ^7-ergostenol and Δ^7-stigmastenol, together with smaller amounts of Δ^5-sterols (e.g. campesterol, stigmasterol, clerosterol and isofucosterol). Rare amino acids, including γ-aminobutyric acid, ethylasparagine, citrulline and cucurbitine (3-aminocarboxypyrrolidine) are also found (*4*, *6*, *14*). Structures of the potential markers, squalene and cucurbitine are presented below.

Cucurbitine



Squalene



Medicinal uses

Uses supported by clinical data

For symptomatic treatment of difficulties with micturition associated with stage I–II prostatic adenoma and irritable bladder (*15–17*).

Uses described in pharmacopoeias and well established documents

No information was found.

Uses described in traditional medicine

Used for the treatment of asthma, burns, constipation, eczema, fever, tapeworms and toothache (*4*, *6*).

Pharmacology

Experimental pharmacology

Anti-androgenic activity

A supercritical carbon dioxide extract of the seeds (dose not stated) antagonized the development of the prostate gland when administered with

testosterone in castrated rats (*4*). A sterol mixture isolated from the seeds dose-dependently inhibited the binding of labelled dihydroxytestosterone to cultured human prostate fibroblasts. Following preincubation of one sample of the fibroblasts with 120 ng of sterol mixture and another with 240 ng of the mixture, dihydroxytestosterone binding was reduced from 68.3% to approximately 47% in the first sample and from 68.3% to 38% in the second (*4*).

Anthelminthic activity

A dried methanol extract of the seeds administered orally to mice, at a dose of 20.0 mg/kg body weight (bw), for 3–4 days had weak activity against *Hymenolepis diminuta*, inducing a 37% clearance of worms in 6 days (*18*). Cucurbitine (3-aminocarboxypyrrolidine) is reported to be one of the most actively anthelminthic constituents of the crude drug (*4*).

Anti-inflammatory activity

The anti-inflammatory effects of intragastric administration of the seed oil to rats with arthritis induced by Freund's complete adjuvant, at a dose of 100 mg/kg bw (*19*), were compared with the effects of indometacin, a classical anti-inflammatory agent. Two models of inflammation were investigated. In the acute inflammatory phase model only seed oil was used, and in the chronic inflammatory phase model both seed oil and indometacin were used. Treatment with the seed oil normalized blood glutathione and serum *N*-acetyl-β-D-glucosaminidase levels, which were elevated in the acute phase of inflammation. Plasma total proteins and albumin, which were reduced during the chronic phase of inflammation, were increased after treatment. Liver glucose-6-phosphate dehydrogenase activity, which was markedly increased during the induction of inflammation, was also reduced after treatment. Also, a remarkable inhibition of paw oedema was observed. A similar pattern was noted following treatment with indometacin (*19*).

Intragastric administration of a supercritical carbon dioxide extract of the crude drug (dose not stated) reduced carrageenan- or dextran-induced oedema in castrated rats (*4*).

Antischistosomal activity

Oral administration of 3.0 g of the seeds daily to mice for 28 days reduced the number of *Schistosoma japonicum* parasites (*20*).

Inhibition of 5α-reductase

The effect of an extract of the seeds on the activity of 5α-reductase, the enzyme responsible for the conversion of testosterone to dihydrotestosterone, was assessed in vitro in cultured human prostate fibroblasts (*4*).

The extract inhibited the activity of the enzyme with a median inhibitory concentration of 128 µg/ml. The affinity of the extract for the dihydrotestosterone receptors was very weak.

Clinical pharmacology

A 12-month randomized, placebo-controlled, multicentre study assessed the efficacy of a 92% ethanol extract of the seeds (15–25:1) in 476 men with benign prostatic hyperplasia stages I and II (mean age 63 years) (*15*). The men were treated with two capsules (500 mg extract per capsule) of the extract (n = 233) or placebo (n = 243) for 12 months. One capsule of the active treatment contained 500 mg pumpkin seed extract (15–25:1, 92% ethanol w/w). The outcome measured was a change in the International Prostate Symptom Score (IPSS). The median baseline IPSS for the placebo group was 17.7 and that for the treatment group was 17.6. A mean reduction of the IPSS of 6.8 in the treatment group and 5.6 in the placebo group (p = 0.014) was reported. A decrease in the IPSS by > 5 points was reported in more patients in the treatment group than in the placebo group, 65% versus 54% (p = 0.021). Urological flow parameters, quality of life, residual urine, prostate volume and prostate-specific antigen remained unchanged in both groups (*15*).

In a 3-month open multicentre study involving 2245 patients with benign prostatic hyperplasia stages I and II according to Alken, the effects of a seed extract (15–25:1, 92% ethanol w/w) were assessed (*22*). Patients were treated with 1–2 capsules of the extract (500 mg per capsule). The results of this study demonstrated an improvement in the IPSS by 41.4% and in quality of life by 46.1%. Micturition (average number of times patients urinated) decreased during the day from 6.7 to 4.8, and at night (nocturia) from 2.7 to 1.1.

In an open study involving 79 male patients with benign prostatic hyperplasia, treatment with the seed fixed oil (dose not specified) reduced the amount of residual urine and led to better bladder emptying and greater volume of emptying after 12 weeks of treatment (*22*).

In a 3-month multicentre open study involving 39 women and 19 men with irritable bladder, patients were treated with 6 g of the crude drug three times daily for 8 weeks. Subjective symptoms such as polyuria and nocturia improved in more than 80% of patients (*17*).

In a study in men, a mixture of Δ^7-sterols, isolated from the crude drug, was administered to patients with benign prostatic hyperplasia. Patients received 90 mg of the mixture by the oral route on days 3 to 4 prior to prostatectomy. A significant decrease in the level of dihydrotestosterone in the prostate tissue was observed (p < 0.05), as well as a significant decrease in serum acid phosphatase (*23*). Oral administration of 30.0 g of the

seed to a single human volunteer decreased urine output, but increased urea and uric acid output over a 3-day period (*24*).

Toxicology
Intraperitoneal administration of an aqueous or aqueous alcoholic extract of the seeds to mice had a median lethal dose of > 5000 mg/kg bw (*25*).

Adverse reactions
Gastrointestinal disorders (heartburn, nausea, stomach ache and diarrhoea), allergic skin reactions and tinnitus.

Contraindications
Hypersensitivity or allergy to the crude drug.
Pregnancy: in traditional medicine, the crude drug has been used as an emmenagogue (*6*), thus extracts of the crude drug should not be ingested during pregnancy.

Warnings
No information was found.

Precautions
No information was found.

Pregnancy: non-teratogenic
See contraindications.

Nursing mothers
Due to the lack of safety data, extracts of the crude drug should not be used during breastfeeding.

Paediatric use
Due to the lack of safety data, extracts of the crude drug should not be used in children aged under 12 years.

Other precautions
No information was found.

Dosage forms
Crude drug and extracts.

Posology

(Unless otherwise indicated)
Oral daily dose: 10 g of seed; equivalent preparations (*16*).

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Folium Cynarae

Definition

Folium Cynarae consists of the dried basal leaves of *Cynara cardunculus* L. (Asteraceae) (*1–4*).

Note: The fresh lower part of the flower head is official in the *African pharmacopoeia* (*5*).

Synonyms

Cynara scolymus L. was the name of the plant cited in the above-mentioned pharmacopoeias and monographs. However, the correct name of the plant is *Cynara cardunculus* L. (Asteraceae) according to the currently accepted nomenclature (*6*).

Selected vernacular names

Alcachofa, alcachofra, alcaucil, alcaucoe, artichaut, artichaut commun, artichiocco, artichoke, artichoke thistle, Artischocke, artiskok, carcioffa, carciofo, carciuffolo, cardo alcachofero, cardo de comer, cardo senzaspine, cardoon, dofital 'roza, edible thistle, enginar, garden artichoke, Gemüseartischocke, globe artichoke, hathi choka, hatichuk, kangar, kangar I dahri, kharshoul, kharsuf, kunjor, Scotch thistle, som-eonggeongqui (*2*, 3, *5*, *7–13*).

Geographical distribution

Native to the Mediterranean, northern Africa and southern Europe, and the Canary Islands; cultivated in subtropical regions (*8*, *14*, *15*).

Description

A large herbaceous perennial, thorny plant, approximately 1.5 m in height. The leaves are large, alternate, deeply dentate. The tall purple flowers are grouped in large capitulums, 10–15 cm in diameter borne by hardy ramified grooved stems, with sessile and almost entire leaves (*5*, *14*, *16*).

Plant material of interest: dried leaves

General appearance

Leaves are very large, up to approximately 50 cm long by 25 cm wide with a long petiole approximately 1 cm thick; lamina deeply pinnatifid, forming flat, lanceolate segments with coarsely-toothed margins; upper surface brownish-green, lower surface greyish-white and densely covered with trichomes; segments with pinnate venation, the side veins terminating in a short point on each marginal tooth; midrib and petiole deeply grooved on the upper surface, the lower surface prominently raised, with several longitudinal ridges and covered with long, whitish trichomes (*1*, *2*, *17*).

Organoleptic properties

Odour: faint, slightly sour; taste: salty at first, then bitter (*1*, *2*, *17*).

Microscopic characteristics

Lamina: The dorsiventral view reveals a fairly large and loosely packed palisade layer of cells. Cells of the upper epidermis have straight to slightly sinuous anticlinal walls, whereas the cells of the lower epidermis are more wavy-walled. Anomocytic stomata on both surfaces, more numerous on the lower surface, with covering trichomes scattered on the upper epidermis, especially over the veins, very abundant on the lower epidermis; individual trichomes mostly of the whiplash type with several small cells forming the uniseriate bases and very long, narrow and sinuous terminal cells intertwining to form a felted mass covering the surface; other less numerous, uniseriate covering trichomes composed of 4–6 cells, tapering to a blunt apex with the cells sometimes more or less globular to ovoid; fairly large glandular trichomes also abundant on both surfaces, with short, 1- or 2-celled stalk and a spherical head filled with a brownish secretion.

Midrib and petiole: Epidermal cells rectangular and longitudinally elongated, with scattered glandular and covering trichomes similar to those in the lamina. Transverse section shows bands of collenchyma below both the upper and lower epidermises; a large vascular bundle in each ridge on the lower surface, and a number of smaller bundles arranged in an arc surrounding the groove on the upper surface; vascular bundles composed of a dense group of pericyclic fibres with thick, lignified walls, a wide area of thin-walled sieve tissue and a lignified xylem containing small vessels, tracheids and xylem parenchyma; below each xylem group a mass of lignified fibres, which, in the larger bundles, extends as a narrow layer on either side of the vascular tissue to join with the fibres of the pericycle; ground tissue composed of large-celled, rounded parenchyma, some with lignified walls (*1*, *2*).

Powdered plant material

Greyish-green to brown powder with faint odour; fragments of the lamina with more or less sinuous walls and anomocytic stomata; covering trichomes, scattered or in felted masses and large, glandular trichomes with brown contents; groups of lignified fibres and vessels from the midrib and petiole, the larger vessels with reticulate thickening (*1*, *2*).

General identity tests

Macroscopic and microscopic examinations (*1*, *2*, *17*), and thin-layer chromatography (*1*, *2*).

Purity tests

Microbiological

Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*18*).

Foreign organic matter

Not more than 2.0% (*1*, *2*).

Total ash

Not more than 15.0% (*1*, *2*).

Acid-insoluble ash

Not more than 4% (*2*).

Water-soluble extractive

Not less than 25.0% (*2*).

Loss on drying

Not more than 8% (*1*).

Pesticide residues

The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*19*). For other pesticides, see the *European pharmacopoeia* (*19*) and the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*18*) and pesticide residues (*20*).

Heavy metals

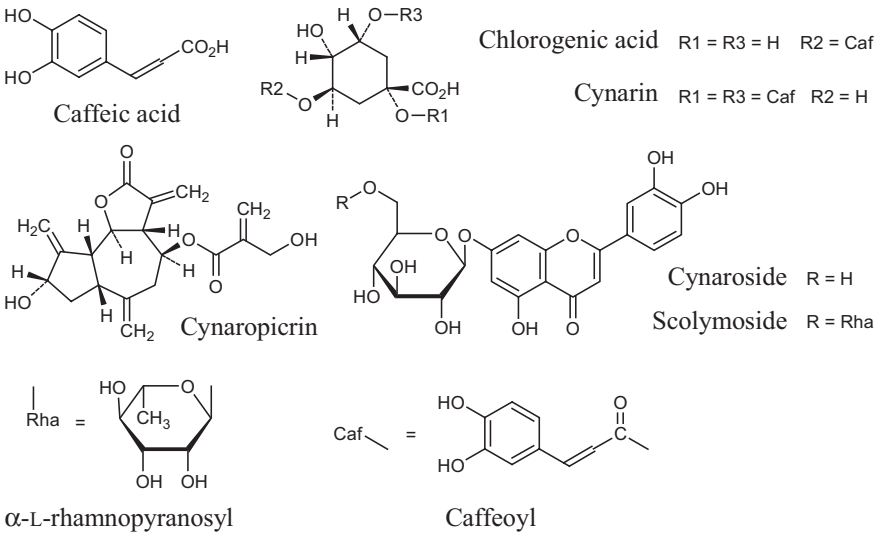
For maximum limits and analysis of heavy metals, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*18*).

Radioactive residues
Where applicable, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*18*).

Other purity tests
Chemical tests to be established in accordance with national requirements.

Chemical assays
To be established in accordance with national requirements.

Major chemical constituents
Contains up to 6% phenolic acids, including 1-*O*-caffeoylquinic acid, 3-*O*-caffeoylquinic acid (chlorogenic acid), caffeic acid, 4-*O*-caffeoylquinic acid, 5-*O*-caffeoylquinic acid, 1,5-di-*O*-caffeoylquinic acid (cynarin); up to 5% sesquiterpene lactones, with cynaropicrin being the primary component, followed by dehydrocynaropicrin, grosheimin and their derivatives; and flavonoids (0.35–0.75%) including scolymoside, cynaroside and cynarotrioside (*7*, *16*, *17*, *21*). The structures of chlorogenic acid, caffeic acid, cynarin, cynaropicrin, scolymoside, cynaroside α-L-rhamnopyranosyl and caffeoyl are presented below.



Medicinal uses
Uses supported by clinical data
Treatment of digestive complaints (e.g. dyspepsia, feeling of fullness, flatulence, nausea, stomach ache and vomiting) (*15*, *22*, *23*). Adjunct treatment of mild to moderate hypercholesterolaemia (*22*, *24–27*).

Uses described in pharmacopoeias and well established documents

Orally for the treatment of atherosclerosis and kidney dysfunctions (diuretic) (*5*).

One study has indicated that the crude drug may be of benefit for the treatment of irritable bowel syndrome (*28*), but further randomized controlled clinical trials are needed before any therapeutic recommendations can be made.

Uses described in traditional medicine

Oral treatment of anaemia, diabetes, fever, gout, rheumatism and urinary stones (*7*, *9*, *29*).

Pharmacology

Experimental pharmacology

Antiatherosclerotic and antihypercholesterolaemic activities

A dried aqueous extract of the leaves (4.5:1) inhibited cholesterol biosynthesis from ^{14}C-acetate in primary cultured rat hepatocytes in a concentration-dependent biphasic manner with moderate inhibition (approximately 20%) being noted between 0.007 and 0.1 mg/ml and stronger inhibition at 1 mg/ml (80%). Replacement of ^{14}C-acetate by ^{14}C-mevalonate largely prevented the inhibitory effects of the extracts, indicating inhibition of the activity of hydroxyl-methyl-glutaryl-CoA-reductase. Stimulation of hydroxyl-methyl-glutaryl-CoA-reductase activity by insulin was efficiently blocked by the extract. Cynaroside and its aglycone luteolin, constituents of the extract, were mainly responsible for enzyme inhibition (*30*). The effect of an extract of the leaves in vivo was investigated in four groups of 10 rats each fed an atherosclerogenic diet. Group one was administered 110 mg/kg body weight (bw) powdered leaves; group two, 80.0 mg/kg bw powdered *Cynara cardunculus*; group three, 10.0 mg/kg bw heparaxal; and group four served as the control. Examination of tissue after 120 days showed that the leaf extract prevented formation of atherosclerotic changes, prevented serum cholesterol increase, caused a decrease in lipid phosphate, slightly increased the level of glycoproteins in the blood, prevented an increase in serum γ-globulin, decreased albumin, glycoproteins and liver cholesterol, and increased γ-globulin and γ-globulin fractions. *Cynara cardunculus* showed a similar but weaker activity (*31*). A methanol extract of the leaves was shown to reduce serum triglyceride levels in olive oil-loaded mice. Oral administration of the extract, at doses between 125 and 500.0 mg/kg bw, significantly suppressed serum triglyceride elevation 2 h after administration of olive oil. In contrast, 6 h after administration of olive oil, increases in triglyceride level were observed in the groups that

received the extract at doses of 125.0 and 250.0 mg/kg bw. Orlistat, a lipase inhibitor, completely suppressed the serum triglyceride elevation at 250.0 mg/kg bw. Clofibrate, a hypolipidaemic medicine, also suppressed the triglyceride level at doses of 250.0 and 500.0 mg/kg bw. Three sesquiterpenes (cynaropicrin, aguerin B and grosheimin) from the extract were isolated as the active components (*32*).

Antihepatotoxic activity

The effects of an aqueous extract of the leaves on taurolithocholate-induced cholestatic bile canalicular membrane distortions were studied in primary cultured rat hepatocytes using electron microscopy. Artichoke extracts at concentrations between 0.08 and 0.5 mg/ml were able to prevent the formation of canalicular membrane transformations in a dose-dependent manner when added simultaneously with the bile acid. However, prevention also occurred when the hepatocytes were preincubated with the extracts, indicating that absorption of the bile acid to components of the extracts was not involved (*33*).

The hepatoprotective activity of cynarin against carbon tetrachloride (CCl_4)-induced toxicity in isolated rat hepatocytes was compared with other phenolic compounds. Only cynarin and, to a lesser extent, caffeic acid showed a cytoprotective effect (*34*). Treatment of rats with three consecutive doses of 500.0 mg/kg bw of an extract of the crude drug, administered by gavage 48, 24 and 1 h before CCl_4 intoxication, produced a significant decrease in glutamic-oxaloacetic transaminase, glutamic-pyruvic transaminase (also known as alanine aminotransferase or ALT), direct bilirubin and glutathione levels, thus indicating a reduction in the potential for hepatotoxicity (*35*).

Primary cultures of rat hepatocytes exposed to *tert*-butyl hydroperoxide were used for characterizing the antioxidative and hepatoprotective potential of an aqueous extract of the crude drug and some selected constituents. Addition of *tert*-butyl hydroperoxide to the culture media resulted in enhanced lipid peroxidation as measured by the production of malondialdehyde and enhanced cytotoxicity detected by leakage of lactate dehydrogenase. The extract added prior to or simultaneously with *tert*-butyl hydroperoxide reduced both phenomena with a median effective concentration (EC_{50}) of 95.0 and 12.0 µg leaf powder/ml, respectively. Furthermore, the aqueous extract prevented the loss of intracellular glutathione caused by *tert*-butyl hydroperoxide. Several polyphenolic and flavonoid constituents of the extract were found to reduce malondialdehyde production. The median effective concentration values were 8.1, 12.5, 15.2 and 28 µg/ml for caffeic acid, chlorogenic acid, cynarin and cynaroside, respectively (*36*).

Primary rat hepatocyte cultures exposed to *tert*-butyl hydroperoxide or cumene hydroperoxide were used to assess the antioxidative and protective potential of aqueous extracts of the leaves. Both hydroperoxides stimulated the production of malondialdehyde, particularly when the cells were pretreated with diethylmaleate in order to diminish the level of cellular glutathione. Addition of the extract did not affect basal malondialdehyde production, but prevented the hydroperoxide-induced increase of malondialdehyde formation in a concentration-dependent manner when presented simultaneously with or prior to the peroxides. The effective concentrations were as low as 0.001 mg/ml (*37*).

Antioxidant activity

A study measured the effects of aqueous and ethanol extracts of the leaves on intracellular oxidative stress stimulated by inflammatory mediators, tumour necrosis factor alpha and oxidized low-density lipoprotein (ox-LDL) in endothelial cells and monocytes. Both extracts inhibited basal and stimulated reactive oxygen species production in endothelial cells and monocytes, in a dose-dependent manner. In endothelial cells, the ethanol extract (50.0 µg/ml) significantly reduced ox-LDL-induced intracellular reactive oxygen species production by 60% ($p < 0.001$) and the aqueous extract (50 µg/ml) reduced ox-LDL-induced intracellular reactive oxygen species production by 43% ($p < 0.01$). The ethanol extract (50 µg/ml) reduced ox-LDL-induced intracellular reactive oxygen species production in monocytes by 76% ($p < 0.01$). Effective concentrations of 25–100 µg/ml were well below the cytotoxic levels of the extracts which started at 1.0 mg/ml as assessed by lactate dehydrogenase leakage and trypan blue exclusion (*38*).

An aqueous dried extract (9:2) of the leaves was studied in human leukocytes to assess activity against oxidative stress. The extract (median effective concentration 0.23 µg/ml) produced a concentration-dependent inhibition of oxidative stress when cells were stimulated with agents that generate reactive oxygen species: hydrogen peroxide, phorbol-12-myristate-13-acetate and *N*-formyl-methionyl-leucyl-phenylalanine. Cynarin, caffeic acid, chlorogenic acid and luteolin, constituents of artichoke leaf extracts, also showed a concentration-dependent inhibitory activity in the above models, contributing to the antioxidant activity of the extract in human neutrophils (*39*).

Choleretic effects

Two aqueous alcoholic extracts of the fresh leaves (total extract containing 19% caffeoylquinic acids, at a dose of 200.0 mg/kg bw and a semi-purified extract containing 46% caffeoylquinic acids, at a dose of 25.0 mg/kg bw) were assessed in rats. Intraperitoneal administration stimulated

choleresis, and significantly increased bile dry residue and total cholate secretion ($p < 0.05$). Intragastric administration of the same extracts (400.0 mg/kg bw, total extract and 200.0 mg/kg bw of the semipurified extract) also increased gastrointestinal motility by 11% and 14%, respectively ($p < 0.05$) (*40*).

The effects of an extract of the crude drug on bile flow and the formation of bile compounds in anaesthetized rats after acute administration and repeated oral administration (twice a day for 7 consecutive days) were studied. A significant increase in bile flow was observed after acute treatment with the extract as well as after repeated administration. The choleretic effects of the extract were similar to those of the reference compound dehydrocholic acid. Total bile acids, cholesterol and phospholipid were determined by enzymatic assays. At the highest dose (400.0 mg/kg bw), a significant increase was observed after single and repeated administration ($p < 0.01$) (*41*).

The choleretic effects of four extracts of the leaves (not described) were assessed in vivo in a study in rats. Extracts 1, 2 and 4 did not show significant choleretic activity at a dose of 1.0 and 2.0 g/kg bw. Extract 3, however, was found to induce an increase of bile flow, which was gradual and sustained. Cynarin and chlorogenic acid, administered as pure compounds, did not show choleretic activity at any of the doses tested and neither of them decreased the malondialdehyde content in liver (*42*).

Toxicology

The oral and intraperitoneal median lethal doses of a hydroalcoholic extract of the leaves in rats were 2.0 g/kg and 1.0 g/kg bw, respectively (*40*). The oral median lethal dose of cynarin in mice was 1.9 g/kg bw. Intraperitoneal administration of cynarin to rats for 15 days, at doses between 50.0 and 400.0 mg/kg bw per day produced no macroscopic, haematological or histological abnormalities. Intraperitoneal administration of cynarin to rats for 40 days at a dose between 100.0 and 400.0 mg/kg bw per day increased body and kidney weight, as well as producing some degenerative changes in the liver (*43*). External application of a leaf extract to the skin of white rats, at doses of 1.0–3.0 g/kg bw for 21 days, did not produce any toxic effects or have any cumulative effects on haematological parameters or the biochemistry of rats. No skin-irritating or eye-irritating effects were observed in guinea-pigs (*44*).

Clinical pharmacology

Antidyspeptic effects

A multicentre open study assessed the effects of a dried aqueous leaf extract (3.8–5.5:1, 320 mg per capsule) in 553 patients with dyspeptic com-

plaints. The daily dose was 4–6 capsules (containing 320.0 mg of extract per capsule) per day, for an average of 43.5 days. Digestive complaints declined significantly, by 71%, over the treatment period ($p < 0.001$). Compared with the baseline data on subjective symptoms, a reduction in abdominal pain (76%), emesis (88%), meteorism (66%) and nausea (82%) was observed. In a subgroup of 302 patients, total cholesterol decreased by 11.5% and triglycerides by 12.5% (*25*). In a similar study, the same extract was assessed in a 6-month open trial involving 203 patients with dyspepsia. The daily dose administered was 3–6 capsules (each capsule containing 320.0 mg of the extract). After 21 weeks of treatment, symptoms such as vomiting, abdominal pain, nausea and flatulence decreased by 84%, 78%, 77% and 70%, respectively. Total blood cholesterol and triglycerides were reduced by 10.9% and 11%, respectively. Data from 159 patients indicated that low-density lipoprotein-cholesterol decreased by 15.8% and high-density lipoprotein-cholesterol increased by 6.3%. Global efficacy as assessed by physicians was good to excellent in 85.7% of patients. No adverse reactions were reported (*26*).

In a double-blind, randomized controlled trial, 247 patients with functional dyspepsia were treated with either a commercial extract of the crude drug (2 × 320.0 mg of plant extract three times daily) or a placebo. The primary outcome measured was the sum score of the patient's weekly rating of the overall change in dyspeptic symptoms (four-point scale). Secondary variables were the scores for each dyspeptic symptom and the quality of life as assessed by the Nepean Dyspepsia Index. Of the 247 patients enrolled, data from 244 patients (129 given active treatment, 115 given placebo) were suitable for inclusion in the statistical analysis (intention-to-treat). The overall improvement in symptoms over the 6 weeks of treatment was significantly greater in patients treated with the commercial extract than in those treated with the placebo (8.3 ± 4.6 versus 6.7 ± 4.8, $p < 0.01$). Similarly, patients treated with the commercial extract showed significantly greater improvement in the global quality of life scores (Nepean Dyspepsia Index) than the placebo-treated patients (-41.1 ± 47.6 versus -24.8 ± 35.6, $p < 0.01$). The preparation tested was significantly better than the placebo at alleviating symptoms and improving the disease-specific quality of life in patients with functional dyspepsia (*45*).

Antihypercholesterolaemic and lipid-lowering effects

Two randomized controlled clinical trials assessed the effects of a dried aqueous extract of the leaves on cholesterol levels in 187 patients (*24, 27*). The first, a randomized, double-blind, placebo-controlled pilot study involving 44 healthy volunteers, assessed the effect of an extract of the crude drug on cholesterol levels. Patients were randomly assigned to receive

either 640.0 mg of the extract or a placebo three times daily for 12 weeks. No significant effects on serum cholesterol were found. However in subgroup analysis, significant cholesterol-lowering effects were observed in subjects with a total cholesterol level of > 210 mg/dl ($p < 0.022$) (*27*).

The second placebo-controlled study assessed the safety and efficacy of a dried aqueous extract of fresh artichoke (25–35:1). Patients received either 1800 mg of artichoke extract as coated tablets, each containing 450.0 mg extract, or a placebo. Patients ($n = 143$) with hyperlipoproteinaemia – initial total cholesterol of > 7.3 mmol/l (> 280 mg/dl) received 1.8 g of a dried leaf extract per day or the placebo for 6 weeks. Changes in total cholesterol and low-density lipoprotein-cholesterol from baseline to the end of treatment showed a statistically significant superiority of the dry artichoke extract over the placebo ($p = 0.0001$). Observed reductions in total cholesterol levels were 18.5% in those who received the extract and 8.6% in those who received the placebo after 6 weeks of treatment (*24*). The decrease in low-density lipoprotein-cholesterol in the group treated with the extract was 22.9% and was 63% in those treated with the placebo. The ratio of low-density lipoprotein to high-density lipoprotein showed a decrease of 20.2% in the group that received the extract and 7.2% in the group that received the placebo. No drug-related adverse events were reported (*24*).

In a randomized, placebo-controlled clinical trial, two groups of 30 patients presenting various dislipidaemic profiles were treated for 50 days with either cynarin, 2 × 250 mg tablets per day, or a placebo. Cynarin was able to induce a significant reduction of hypercholesterolaemia ($p < 0.001$), the level of pre-β-lipoproteins ($p < 0.01$), the β/α-lipoprotein ratio ($p < 0.01$) and patient's body weight (*46*).

Several uncontrolled studies have found that cynarin reduced total serum cholesterol in patients after treatment with oral doses of 750–1500 mg per day. Oral administration of cynarin to 17 patients, at a dose of 1000 mg/day, for 4 weeks resulted in a significant decrease in total cholesterol (15%, $p < 0.005$) (*9*).

Choleretic effect

A randomized, double-blind, placebo-controlled trial assessed the choleretic effects of a dry aqueous extract of the leaves (4.5–5:1) in 20 male volunteers with acute or chronic metabolic disorders. The treatment group ($n = 10$) received a single intraduodenal dose of the extract at a dose of 1.92 g/day in 50 ml of water on an empty stomach, while the control group received a placebo of similar appearance. Crossover to the alternative treatment followed an 8-day washout period. The outcomes included intraduodenal bile secretion measured using multi-channel probes. Com-

pared with baseline values, 60 minutes after administration there was a significant increase in bile secretion in the treatment group (151%) as compared with the placebo group ($p < 0.01$) (*23*).

Irritable bowel syndrome

Irritable bowel syndrome, characterized by abdominal pain and altered bowel habit, has symptoms that overlap with those of dyspepsia. Since the crude drug is used for the treatment of dyspepsia, a postmarketing surveillance study was performed to assess its effects on irritable bowel syndrome. A subgroup of patients ($n = 279$) with symptoms of irritable bowel syndrome was identified from a sample of individuals ($n = 553$) with dyspeptic syndrome who were being monitored in a postmarketing surveillance study of the extract for 6 weeks. Analysis of the data from the subgroup with irritable bowel syndrome revealed significant reductions in the severity of symptoms including abdominal pain, bloating, flatulence and constipation, and favourable evaluations of overall effectiveness by both physicians and patients (*28*).

Pharmacokinetics

A study to investigate the absorption, metabolism and disposition of artichoke leaf extract was performed using two different extracts (*47*). The extracts were administered to 14 healthy volunteers in a crossover study. Each subject received doses of both extracts. The administered dose of extract A contained caffeoylquinic acids equivalent to 107.0 mg caffeic acid and luteolin glycosides equivalent to 14.4 mg luteolin. The administered dose of extract B contained caffeoylquinic acids equivalent to 153.8 mg caffeic acid and luteolin glycosides equivalent to 35.2 mg luteolin. Urine and plasma analysis were performed by a validated high-performance liquid chromatography method using 12-channel coulometric array detection. None of the genuine target extract constituents could be detected in the plasma or urine of the subjects. However, caffeic acid, its methylated derivates ferulic acid and isoferulic acid and the hydrogenation products dihydrocaffeic acid and dihydroferulic acid were identified as metabolites derived from caffeoylquinic acids. Except for dihydroferulic acid, all of these compounds were present as sulfates or glucuronides. Peak plasma concentrations of total caffeic acid, ferulic acid and isoferulic acid were reached within 1 h and declined over 24 h showing almost biphasic profiles. By contrast, maximum concentrations for total dihydrocaffeic acid and dihydroferulic acid were observed only after 6–7 h, indicating two different metabolic pathways for caffeoylquinic acids. Luteolin administered as glucoside was recovered from plasma and urine only as sulfate or glucuronide, but neither in the form of genuine glucosides nor

as free luteolin. Peak plasma concentrations were reached rapidly within 0.5 h. The elimination showed a biphasic profile (*47*).

Adverse reactions

Gastrointestinal complaints included mild diarrhoea, accompanied by abdominal cramps, upper abdominal pain, nausea and heartburn. Allergic reactions may occur in sensitized patients (*22, 25*).

No significant adverse events other than gastrointestinal discomfort have been reported from open or controlled clinical trials (*24–27*, *48*).

Contraindications

Hypersensitivity or allergies to artichokes and other plants from the Compositae/Asteraceae, and obstruction of the bile ducts (*15*).

Warnings

Possible interaction with coumarin-type anticoagulants.

Precautions

General

Patients with gallstones should seek the advice of a health care provider prior to use.

Carcinogenesis, mutagenesis, impairment of fertility

The genotoxic effects of flavonoid constituents present in the crude drug (quercetin and luteolin) were assessed in two short-term bacterial assays (*49*). In *Salmonella typhimurium* (strains TA1538 uvrB- and TA1978 uvrB+) the flavonoids did not induce damage in the DNA as recognized by UvrABC nuclease. Results of the SOS-chromotest in *Escherichia coli* K-12 strains PQ37 and PQ243 indicated that the flavonoids only weakly induced the SOS system (*49*).

Drug interactions

No information was found.[1]

Pregnancy: teratogenic effects

Due to the lack of safety and efficacy studies, the use of the crude drug during pregnancy is not recommended.

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[1] A report of a potential drug interaction with Folium Cynarae or its preparations and with coumarin-type anticoagulants such as phenprocoumone and warfarin has been recorded by a national regulatory authority.

Pregnancy: non-teratogenic effects
Due to the lack of safety data, the use of the crude drug during pregnancy is not recommended.

Nursing mothers
Due to the lack of safety data, the use of the crude drug during breastfeeding is not recommended.

Paediatric use
Due to the lack of safety data, the use of the crude drug for the treatment of children under the age of 12 years is not recommended.

Other precautions
No information was found.

Dosage forms
Crude drug, extracts and other Galenical preparations for internal use.

Posology
(Unless otherwise indicated)
Average oral daily dose: for hypercholesterolaemia and dyspepsia, 1–2 g of a dried aqueous extract (*24*, *27*, *45*). Adult daily dose: 5–10 g of crude drug; or equivalent preparations (*15*, *17*).

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Cortex Granati

Definition

Cortex Granati consists of the dried root or trunk bark of *Punica granatum* L. (Lythraceae) (*1–3*).

Synonyms

Punica nana L. (Punicaceae) (*4*).

Note: According to current botanical authorities, *Punica granatum* belongs to the Lythraceae family (*5*).

Selected vernacular names

Anar, anara, anar-ke-per, Carthaginian apple, dadam, dadima-phalam, dalima, dalimb, dalimbay, dalimbuhannu, dalimo, darakte-naiar, darimba, darinko bokra, daru, delima, delum, delun, dhalima, dila dae lok, dlima, ende limau, gangsalan, glima glineu mekah, granada, granade, granado, Granatbaum, granatum, grenadier, grenadillo, gul armini, gulnar, jaman, jeliman, kupchaphala, lelo kase, madalai, madalam, madalangkai, matalam, mathalanarkom, melograno, mkoma manga, nar, pomegranate, posnar, qsur roman, qsur rommam, quishr-al-romman, quishr-romman, ranato, romã, roman, romeira, rommam, roman amruj, romanzeira, roumammam-goulnar, ruman, rumau, sekiryuu-karpi, seog-ryu, seokryupi, shajratur-rummam, sham-al-rumman, shih liu pi, shiliupi, shukadana, talima, thab thim, thap thim, zakuro-juhi (*1*, *3*, *4*, *6–12*).

Geographical distribution

Native to the Middle East (eastern Mediterranean to northern India), and now widely cultivated in warm regions throughout the world (*6*, *8*, *9*).

Description

A deciduous shrub or small tree, erect, up to 7 m high, much branched from the base; branches slender; branchlets often ending in spines, the young ones quadrangular or almost tetrapterous. Leaves: simple, oppo-

site, verticillate, oblong-lanceolate, glabrous, 1–9 cm long and 0.5–2.5 cm wide; apex acute, obtuse or emarginate; base cuneate. Flowers: 1–5 at the highest leaf axil of branchlets, 1 terminal, sessile or subsessile. Calyx: 2–3 cm long, red or pale yellow; lobes erectopatent to recurved; petals round or obtuse, red or white. Fruit, a berry, globose, 5–13 cm in diameter, with a leathery rib enclosing numerous seeds, variously coloured, yellowish-green, white, reddish brown or rarely blackish-purple. Seeds: numerous, red, pink or yellowish white (*6*, *7*, *9*).

Plant material of interest: root and stem bark

General appearance

Stem bark: curved pieces or quills; up to 15 cm long, 0.5–3.0 mm thick: outer surface, yellowish to blackish-brown, with occasional greyish patches of lichens, longitudinally wrinkled and marked; small, broadly elliptical lenticels; inner surface, light yellow to light brown, finely striated; fracture, very short and granular.

Root bark: flat, irregular, curved, or recurved small pieces; outer surface, brownish yellow, rough, with darker patches and conchoidal depressions due to exfoliation of the outer portion, but no lenticels; inner surface, yellow, smooth, with irregular darker brown patches. Other characteristics similar to those of stem bark (*1–3*).

Organoleptic properties

Odour: slight; taste: astringent and slightly bitter (*1*, *2*).

Microscopic characteristics

Cork, formed of several alternating layers of suberized thin-walled cells and of lignified cells with greatly thickened inner tangential walls. Cortex, consisting of parenchyma containing small starch granules; crystals of calcium oxalate from scattered prisms to rosette clusters; and large sc1ereids, which are isolated, rarely in small groups, with very thick and strongly stratified walls, up to 400 µm long and 200 µm broad. Phloem shows numerous cells containing cluster crystals of calcium oxalate in more or less tangential rows, and parenchyma cells with numerous small starch granules or amorphous tannin masses. Medullary rays, 1–2 cell rows with occasional cells containing numerous small prisms (*1–3*).

Powdered plant material

Yellowish brown to dark brown, characterized by fragments of parenchyma containing numerous starch granules and crystals of calcium oxalate; sclereids with very thick and pitted walls; fragments of cork with

prominent, thickened and lignified walls; numerous calcium oxalate crystals, prisms, 6–10 µm long and rosette cluster crystals up to 15 µm in diameter; starch granules, abundant, simple, 2–10 µm in diameter, or rarely compound; occasional long wood fibres, 15–20 µm in diameter, associated with pitted vessels; bast fibres absent (*1*, *3*).

General identity tests

Macroscopic and microscopic examinations, microchemical tests (*1–3*, *13*) and thin-layer chromatography (*3*, *13*).

Purity tests

Microbiological

Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*14*).

Foreign organic matter

Not more than 2% (*1*, *2*).

Total ash

Not more than 15% (*2*).

Acid-insoluble ash

To be established in accordance with national requirements.

Water-soluble extractive

To be established in accordance with national requirements.

Alcohol-soluble extractive

To be established in accordance with national requirements.

Loss on drying

To be established in accordance with national requirements.

Pesticide residues

The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*15*). For other pesticides, see the *European Pharmacopoeia* (*15*) and the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*14*) and pesticide residues (*16*).

Heavy metals

For maximum limits and analysis of heavy metals, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*14*).

Radioactive residues

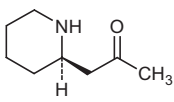
Where applicable, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*14*).

Chemical assays

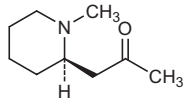
Contains not less than 0.4% of total alkaloids.

Major chemical constituents

The major biologically active constituents are alkaloids and tannins. Isopelletierine, methylpelletierine, methylisopelletierine, pseudopelletierine, norpseudopelletierine and related alkaloids totalling 0.5–0.9%. The tannins present at up to 22% in the bark include punicalin, its 2-*O*-galloyl derivative and punicalagin (*12*, *17*, *18*). The structures of pelletierine, *N*-methylisopelletierine, pseudopelletierine, and the tannins punicalin and punicalagin are presented below.

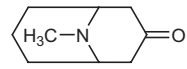


Pelletierine



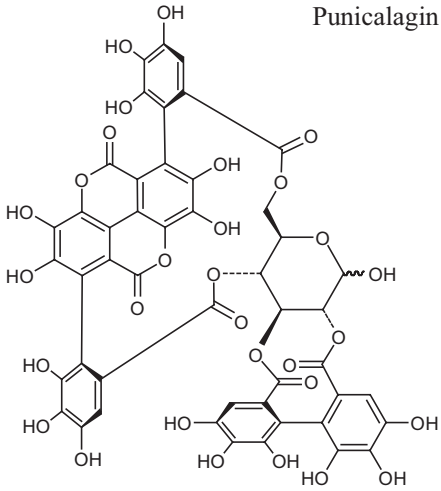
and enantiomer

N-Methylisopelletierine

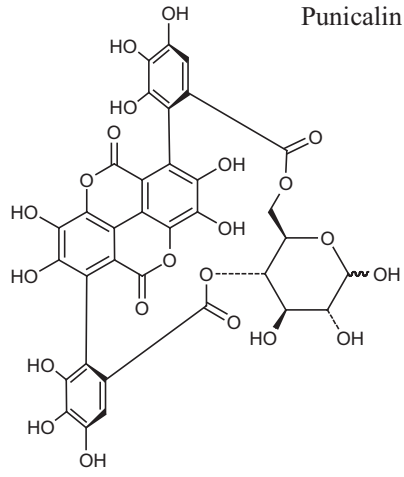


Pseudopelletierine

Punicalagin



Punicalin



Medicinal uses

Uses supported by clinical data

No information was found.

Uses described in pharmacopoeias and well established documents

Used orally for the treatment of diarrhoea and intestinal parasites (*7*, *19*).

Uses described in traditional medicine

Used orally to treat dyspepsia, sore throat, menorrhagia, leukorrhoea and ulcers (*12*).

Pharmacology

Experimental pharmacology

Antimicrobial activity

An aqueous and a 95% ethanol extract of the bark had weak activity in vitro against *Bacillus cereus*, *Enterococcus faecalis*, *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa* and *Staphylococcus aureus* at a concentration of 25.0 mg/well (*20*). A decoction of the crude drug inhibited the growth of *Trichophyton tonsurans, T. rubrum*, *T. simii*, *Trichosporon beigelii*, *Microsporum fulvum, M. gypseum* and *Candida albicans* when added to the nutrient medium at a concentration of 5% (*21*).

Anthelminthic and molluscicidal activities

Pelletierine, an alkaloid constituent of the bark, was active against tapeworms (*Taenia solium*), but was not active against other intestinal parasites (*4*). At a concentration of 1:10 000, pelletierine hydrochloride kills tapeworms within 5–10 minutes (*22*). This alkaloid acts by causing the tapeworm to relax its grip on the intestinal walls and thereby making it possible to be expelled by cathartics. The molluscicidal activity of the crude drug against the snail *Lymnaea acuminata* was found to be both time- and dose-dependent. An ethanol extract of the bark was effective in killing the test animals, with a 24 h median lethal concentration of 22.42 mg/l (*23*). The extract was not toxic to the fish, *Colisa fasciatus*, which shares the same habitat with the snail (*23*).

Antiuraemic activity

Administration of a decoction of the bark in the drinking-water, at a dose of approximately 150.0 mg/kg body weight (bw), prevented casein/adenine-induced kidney failure in rats (*24*).

Pharmacokinetics

The metabolism of punicalagin, a water-soluble ellagitannin isolated from the crude drug, was assessed in rats (*25*). The animals were treated with repeated oral administration of a 6% punicalagin-containing diet for 37 days. Punicalagin and related metabolites were identified by high performance liquid chromatography–diode array detector–mass spectrome-

try–mass spectrometry (HPLC-DAD-MS-MS) in plasma, liver and kidneys. Five punicalagin-related metabolites were detected in liver and kidney, that is, two ellagic acid derivatives, gallagic acid, 3,8-dihydroxy-6H-dibenzo[b,d]pyran-6-one glucuronide, and 3,8,10-trihydroxy-6H-dibenzo[b,d]pyran-6-one (*25*).

Toxicology

In animal experiments, intragastric administration of very large doses (not stated) of the alkaloids isolated from the bark caused respiratory arrest and death (*26*). Punicalagin has been reported to be toxic to cattle and rats (*25*). The chronic toxicity of punicalagin was assessed in rats. Treatment consisted of repeated oral administration of a 6% punicalagin-containing diet for 37 days. Feedstuff intake, food utility index and growth rate were lower in treated rats during the first 15 days, but no significant adverse effects were observed. No significant differences were found in treated rats in any blood parameter analysed (including the antioxidant enzymes glutathione peroxidase and superoxide dismutase) with the exception of urea and triglycerides, which remained low throughout the experiment (*25*).

Clinical pharmacology

Toxicology

Ingestion by humans of more than 80.0 g of drug may cause severe vomiting with blood, dizziness, fever, tremor and collapse. After 10 hours to 3 days temporary blindness may occur, which usually resolves after several weeks. Ingestion of pelletierine may cause visual disturbances with mydriasis (dilated pupils), dizziness and headache, as well as long-lasting anaesthesia or somnolence. Further symptoms of overdose include colic, cold sweat, dizziness, headache, muscle cramps, weakness or paralysis of the lower extremities, nausea, cardiac and respiratory collapse (*27*).

Adverse reactions

Common adverse events observed in humans include dizziness, visual disturbances, weakness, calf spasms and tremors. Large overdoses (> 80.0 g) of the crude drug may lead to dizziness, mydriasis, severe headache, vertigo, vomiting, lethargy, collapse and possible death due to the alkaloid content (*26*).

Contraindications

Hypersensitivity or allergy to the bark.

Warnings

For diarrhoea lasting for longer than 3 days, contact a health care provider. For diarrhoea associated with fever, nausea and vomiting, or bloody stools, contact a health care provider. Do not exceed recommended dosage.

Precautions

General

No information was found.

Drug interactions

No information was found.

Drug and laboratory test interactions

No information was found.

Carcinogenesis, mutagenesis, impairment of fertility

The genotoxic effects of the crude drug were examined in established human cell lines, Raji and P3HR-1. Cells were treated with a decoction of the bark at various concentrations for 24 and 48 hours in vitro. Cell growth and viability were dose-dependently reduced. No apparent chromosomal aberrations were induced by the treatment. Administration of a bark extract induced apoptotic DNA fragmentation (*28*).

Pregnancy: teratogenic effects

No information was found.

Pregnancy: non-teratogenic effects

Due to the lack of safety data, the use of Cortex Granati during pregnancy is not recommended.

Nursing mothers

Due to the lack of safety data, the use of Cortex Granati during breastfeeding is not recommended.

Paediatric use

Due to the lack of safety data, the use of Cortex Granati in children under the age of 12 years is not recommended.

Other precautions

No information was found.

Dosage forms

Crude drug, extracts and tablets. Store in a cool dry place (*19*).

Posology

Oral daily dose: 3–9 g for decoction (*19*).
Daily dose: 20 g root bark fluidextract (1:1) in 59% ethanol for the treatment of *Taenia* infestation (*27*).

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Pericarpium Granati

Definition

Pericarpium Granati consists of the dried pericarp of *Punica granatum* L. (Lythraceae) (*1–3*).

Synonyms

Punica nana L. (Punicaceae) (*4*).

Note: According to current botanical authorities, *Punica granatum* belongs to the Lythraceae family (*5*).

Selected vernacular names

Anar, anara, anar-ke-per, Carthaginian apple, dadam, dadima-phalam, dalima, dalimb, dalimbay, dalimbuhannu, dalimo, darakte-naiar, darimba, darinko bokra, daru, delima, delum, delun, dhalima, dila dae lok, dlima, ende limau, gangsalan, glima glineu mekah, granada, granade, granado, Granatbaum, granatum, grenadier, grenadillo, gul armini, gulnar, jaman, jeliman, kupchaphala, lelo kase, madalai, madalam, madalangkai, matalam, mathalanarkom, melograno, mkoma manga, nar, pomegranate, posnar, qsur roman, qsur rommam, quishr-al-romman, quishr-romman, ranato, romã, roman, romeira, rommam, roman amruj, romanzeira, roumammam-goulnar, ruman, rumau, sekiryuu-karpi, seog-ryu, seokryupi, sham-al-rumman, shajratur-rummam, shih liu pi, shiliupi, shukadana, talima, thab thim, thap thim, zakuro-hi (*2–4*, *6–12*).

Geographical distribution

Native to the Middle East (eastern Mediterranean to northern India), and now widely cultivated in warm regions throughout the world (*6*, *8*, *9*).

Description

A deciduous shrub or small tree, erect, up to 7 m high, much branched from the base; branches slender; branchlets often ending in spines, the young ones quadrangular or almost tetrapterous. Leaves: simple, oppo-

site, verticillate, oblong-lanceolate, glabrous, 1–9 cm long and 0.5–2.5 cm wide; apex acute, obtuse or emarginate; base cuneate. Flowers: 1–5 at the highest leaf axil of branchlets, 1 terminal, sessile or subsessile. Calyx 2–3 cm long, red or pale yellow; lobes erectopatent to recurved; petals round or obtuse, red or white. Fruit, a berry, globose, 5–13 cm in diameter, with a leathery rind enclosing numerous seeds, variously coloured, yellowish green, white, reddish brown or rarely blackish purple. Seeds: numerous, red, pink or yellowish white (*6*, *7*, *9*).

Plant material of interest: pericarp

General appearance

Irregular slices or gourd-shaped, brittle, varying in size, 1.5–3.0 mm thick. The outer surface reddish brown, brownish yellow or dark brown; somewhat lustrous, rough, with numerous warty protuberances. Some with a raised tubular persistent calyx and a stout and short peduncle or its scar. Inner surface yellow or reddish brown, with raised reticulated remains of the peduncle. Texture hard and fragile, fracture yellow, somewhat granular (*1*, *6*).

Organoleptic properties

Odourless; taste: bitter and astringent (*1*, *6*).

Microscopic characteristics

Transverse section of the pericarp shows the following: exocarp consists of irregular polygonal cells with slightly thickened but not lignified outer wall and a thick smooth cuticle. Mesocarp generally consists of thin-walled parenchyma cells containing starch granules, clusters or prisms of calcium oxalate. Scattered among the parenchyma cells are sclereids occurring singly or in small groups with strongly thickened lignified walls and rather small lumen or with slightly thickened somewhat lignified walls and wider lumen. Small bicollateral vascular bundles course through the parenchyma; rather thick-walled, non-lignified fibres occasionally accompanying the bundles. Larger bicollateral vascular bundles course through the central part of the mesocarp: phloem fibres are few and possess clear non-lignified walls and a narrow lumen; some phloem parenchyma contains prisms of calcium oxalate. Endocarp cells relatively small, containing starch granules and crystals of calcium oxalate, and stone cells also relatively small (*1*, *6*).

Powdered plant material

Reddish brown. Stone cells sub-rounded, rectangular or irregular, rarely branched, 27–102 μm in diameter, with relatively thick walls and a large lumen, some containing brown matter. Epidermal cells sub-square or sub-

rectangular, with slightly thickened walls. Clusters of calcium oxalate 10–25 µm in diameter, prisms infrequent. Spiral and reticulate vessels 12–18 µm in diameter. Starch granules sub-rounded, 2–10 µm in diameter (*1*).

General identity tests

Macroscopic and microscopic examinations (*1*, *2*, *6*), microchemical tests (*1*, *6*), and thin-layer chromatography (*2*, *6*).

Purity tests

Microbiological

Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*13*).

Foreign organic matter

Not more than 6% (*1*).

Total ash

Not more than 4% (*2*, *6*).

Acid-insoluble ash

Not more than 1% (*6*).

Water-soluble extractive

Pericarp: not less than 30% (*6*).

Alcohol-soluble extractive

Pericarp: not less than 15% (*6*).

Loss on drying

Not more than 10% (*6*).

Pesticide residues

The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*14*). For other pesticides see the *European pharmacopoeia* (*14*) and the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*13*) and pesticide residues (*15*).

Heavy metals

For maximum limits and analysis of heavy metals, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*13*).

Radioactive residues

Where applicable, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*13*).

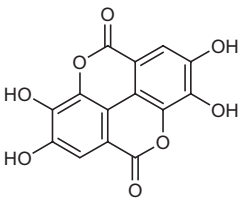
Chemical assays

Total tannins not less than 10% (*1*).

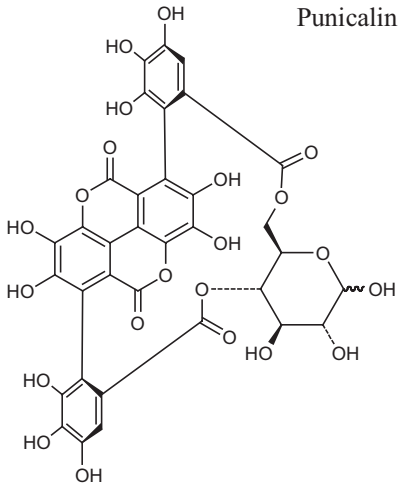
Major chemical constituents

The major constituents are hydrolysable ellagitannins (up to 28%) and other polyphenols (*3*, *12*, *16*, *17*). Structures of ellagic acid and punicalin are presented below.

Ellagic acid



Punicalin



Medicinal uses

Uses supported by clinical data

None.

Uses described in pharmacopoeias and well established documents

Orally for the treatment of chronic diarrhoea, dysentery, gingivitis and intestinal parasites (*1*, *7*, *18*).

Uses described in traditional medicine

Treatment of bronchitis, fever, gastrointestinal ailments, menorrhagia, respiratory tract infections, skin rashes, vaginal infections and worms (*12*, *19*).

Pharmacology

Experimental pharmacology

Antidiarrhoeal activity

Intragastric administration of a decoction or a 95% ethanol extract of the crude drug at a dose of 200.0 mg/kg or 50.0 mg/kg body weight (bw), respectively, reduced faecal output in rats with castor-oil induced diarrhoea (*21*). The same extracts, at a dose of 500.0 mg/kg bw, exhibited intestinal antisecretory activity in rats with magnesium sulfate-induced enteropooling (*20*).

Antimicrobial, antiparasitic and antiviral activity

An aqueous, butanol, or 95% ethanol extract of the crude drug had in vitro activity against *Proteus mirabilis*, *Haemophilus influenzae*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, *Streptococcus aureus*, *Staphylococcus aureus* and *Candida albicans* at a concentration of 60.0 µg/ml (*21*, *22*). An aqueous extract and a 95% ethanol extract of the fruit rind had weak in vitro activity against *Bacillus cereus*, *Enterococcus faecalis*, *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa* and *Staphylococcus aureus* at a concentration of 25 mg/well (*23*, *24*).

A decoction of the crude drug weakly inhibited the growth of *Trichophyton tonsurans, T. rubrum*, *T. simii, Trichosporon beigelii*, *Microsporum fulvum*, *M. gypseum* and *Candida albicans* when added to the nutrient medium at a concentration of 5% (*25*).

Tannins separated from the crude drug were effective against genital herpes virus (herpes simplex virus type 2). The tannin not only inhibited replication of the virus, but also blocked herpes type 2 viral adsorption in cultured cells (*26*).

An aqueous extract of the crude drug had antiviral activity against hepatitis B virus in PLC/PRF/5 cells in vitro (*27*). An aqueous extract inhibited hepatitis C virus in vitro at a concentration of 100 µg/ml. An acetone extract of the crude drug had larvicidal activity against *Chrysomya albiceps* at a concentration of 25 µg/ml (*28*). At a concentration of 10.0 ml/plate, an aqueous extract of the crude drug weakly inhibited the growth of *Ascaris galli, Pheritima posthuma* and *Taenia solium* (*29*), as well as *Ascaris lumbricoides* (*30*).

Antioxidant activity

Aqueous, alcohol and ethyl acetate extracts of the crude drug have shown significant antioxidant activity in various in vitro and in vivo models. A methanol extract of the crude drug had antioxidant activity in vitro, in the β-carotene-linoleate and 1,1-diphenyl-2-picrylhydrazyl radical model

systems. The methanol extract exhibited 83% and 81% antioxidant activity at a concentration of 50.0 µg/ml using the β-carotene-linoleate and 1,1-diphenyl-2-picrylhydrazyl radical model systems, respectively (*31*). A dried methanol extract fed to rats (50.0 mg/kg bw) following treatment with carbon tetrachloride (CCl_4) (2.0 g/kg bw) decreased the levels of catalase, superoxide dismutase and peroxidase by 81, 49 and 89%, respectively. Pretreatment of the rats with the methanol extract maintained the levels of catalase, peroxidase and superoxide dismutase at values comparable with control values, whereas lipid peroxidation was reduced by 54% compared to the control group (*32*).

Antiulcer activity

Intragastric administration of an aqueous extract (5 ml/kg bw) or an unspecified protein-containing fraction (50 mg/kg bw) of the crude drug to rats prevented hydrochloric acid and ethanol-induced gastric ulceration (*33*). The gastroprotective effects of tannic acid and the aqueous extract of the crude drug against ethanol-induced damage were investigated in rats. Oral administration of the extract or tannic acid induced a significant decrease in gastric lesions (48–76%). The protection observed was more pronounced when the test solution was given at the same time as the ethanol. The acid content of the stomach was increased by 368% after administration of the ethanol extract (*34*).

Immune effects

Oral administration of a powder made from the fruit rind, at a dose of 100.0 mg/kg bw, as an aqueous suspension, stimulated cell-mediated and humoral components of the immune system in rabbits. The suspension elicited an increase in antibody titre to typhoid-H antigen. It also enhanced the inhibition of leukocyte migration in the leukocyte migration inhibition test and induration of skin in the delayed hypersensitivity test with purified protein derivative, confirming its stimulatory effect on the cell-mediated immune response (*35*).

Toxicology

An aqueous extract of the crude drug was reported to stimulate uterine contractions in non-pregnant rats but the dose and the route of administration were not stated (*36*).

Clinical pharmacology

A water-soluble component derived from an ethanol extract of the crude drug possessing antimicrobial activity was used in an antibacterial mouthwash at a concentration of 0.8% w/w (*18*). The mouthwash was composed of a combination of the water-soluble component (0.8 g/100 ml),

menthol (0.03 g/100 ml), and absolute alcohol (5 g/100 ml). Eighty volunteers (30 healthy and 50 with dental caries) were required to rinse their mouths with 20 ml of the mouthwash for 30 seconds, they then closed their mouths for 90 seconds and then spat out their saliva for analysis. The salivary microbes were cultured and identified. Oral pathogens such as *Staphylococcus aureus*, *S. mutans* and *Lactobacillus* spp. were killed whereas *Candida albicans* was not (*18*).

Adverse reactions

No information was found.

Contraindications

Hypersensitivity or allergy to the plant material.

Warnings

In cases of diarrhoea lasting for longer than 3 days, or associated with fever, nausea and vomiting, or bloody stools, contact a health care provider. Do not exceed recommended dose.

Precautions

Carcinogenesis, mutagenesis, impairment of fertility

The genotoxic effects of the crude drug were examined in established human cell lines, Raji and P3HR-1. Cells were treated with a decoction of the crude drug at various concentrations for 24 and 48 hours in vitro. Cell growth and viability were dose-dependently reduced. No apparent chromosomal aberrations were induced by the treatment. Administration of the extract induced apoptotic DNA fragmentation (*37*).

Pregnancy: teratogenic effects

No information was found.

Pregnancy: non-teratogenic effects

Due to the lack of safety data, the use of the crude drug during pregnancy is not recommended.

Nursing mothers

Due to the lack of safety and efficacy studies, the use of the crude drug by breastfeeding mothers is not recommended.

Paediatric use

Due to the lack of safety data, the use of the crude drug for the treatment of children under the age of 12 years is not recommended.

Other precautions
No information was found.

Dosage forms
Crude drug, extracts and tablets. Store in a cool dry place (*1*).

Posology
(Unless otherwise indicated)
Oral daily dose: 2.5–4.5 g (*1*, *7*).

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Folium Guavae

Definition
Folium Guavae consists of the dried and/or young leaves of *Psidium guajava* L. (Myrtaceae) (*1*).

Synonyms
Psidium aromaticum L. *P. cujavillus* Burm. f, *P. pomiferum* L., *P. pyriferum* L., *P. pumilum* Vahl (*2*).

Selected vernacular names
Abas, aduoba, aguoba, amba, amrood, amrud, amrut, amruta-phalam, araca-goiba, arasa, banjiro, banziro, bidji, bihi, bilauti, borimak, bugoyab, buyaki, dijamboé, coloc, djambu bidji, djambu klutuk, eguabe, fa-rang, goavy, goejaba, goiaba, goiabeira, goiabeira-vermelha, goiabeiro, gouyav, gouyavier, goyav, goyavier, goyya, grosse gelbe, gua, guafa, guajave, guava, guava tree, Guave, guayaba, guayaba cak, guayaba colorada, guayaba cotorrera, guayaba de gusano, guayaba de venado, guayaba del Peru, guayaba peruana, guayabe, guayabero, guayabo, guayabo agrio, guayabo blanco, guayava, Guayave, guega, guyaba, gwaabaa, gwawa, hind armudu, ipera, jaama, jamba, jambu biji, kautonga, Kiswahili, koyya, krue, ku'ava, kuabas, kuava, kuawa, kuiaba, kuliabas, mabera, maduriam, manssla, motiram, mpera, mugwavha, ngoaba, nulu, oguawa, pat'a, perala, pero delle Indie, peyara, posh, psidio, psidium, punjo, quwawa, sari guafa, sigra, sikra, tuava, warakel-guafa, wariafa, woba, xalxoctl (*1*, *3–10*).

Geographical distribution
Native to tropical America, but now pantropical (*7*, *8*, *11*).

Description
A large shrub or small tree up to 10 m high. Stem slender, usually not exceeding 30 cm in width; bark brownish, thin, smooth, and often flaking off in scaly patches. Leaves opposite, oblong, slightly oval shaped, 5–15 cm in length, light green on the upper surface, and downy and pale green on

the underside. They display prominent veins on the underside of the leaf. Flowers white, occur either singly or a few in a cluster in axils of the leaves. About 2.5 cm in diameter, with numerous stamens arranged in groups. Fruits are globose or pyriform, yellow, usually 2.5–10 cm long and 2.5–5 cm in diameter, horticulture varieties with larger fruits with an edible pink mesocarp are common. They contain many seeds, and the calyx persists in fruit (*7*, *8*).

Plant material of interest: dried or young leaves

General appearance

Oblong, slightly oval, apex acute, round or acuminate; base symmetrical, cordate; margin smooth or dentate; venation reticulate; green (*1*).

Organoleptic properties

Odour: slight; taste: astringent (*1*).

Microscopic characteristics

The surface view shows nearly straight anticlinal epidermal cell walls on both surfaces, those on upper surface thickened; abundant stomata and covering trichomes on lower surface; oil glands on both surfaces, but more frequent on lower surface. Transverse section shows small epidermal cells with straight anticlinal walls, upper epidermal layer 2–3 cells thick with a few oil glands (schizogeneous); palisade tissue multi-layered; numerous oil glands in the mesophyll region; midrib region shows collenchymatous cells; bicollateral vascular bundle is distinctly horse-shoe shaped and surrounded by lignified pericyclic fibres; xylem elements are generally lignified; uniseriate trichomes smooth-walled and abundant on lower epidermis (*1*).

Powdered plant material

Light green; taste astringent; lamina fragments show abundant stomata, punctate, numerous covering trichomes; rosette crystals; lignified vascular elements (*1*).

General identity tests

Macroscopic and microscopic examinations (*1*).

Purity tests

Microbiological

Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*12*).

Foreign organic matter
To be established in accordance with national requirements.

Total ash
To be established in accordance with national requirements.

Acid-insoluble ash
To be established in accordance with national requirements.

Water-soluble extractive
To be established in accordance with national requirements.

Loss on drying
To be established in accordance with national requirements.

Pesticide residues
The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*13*). For other pesticides, see the *European pharmacopoeia* (*13*) and the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*12*) and pesticide residues (*14*).

Heavy metals
For maximum limits and analysis of heavy metals, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*12*).

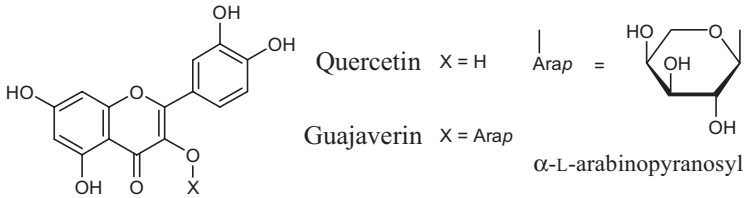
Radioactive residues
Where applicable, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*12*).

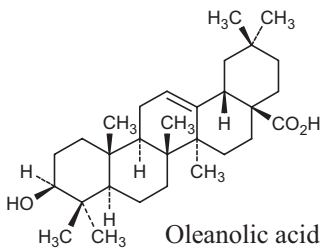
Chemical assays
Not less than 14% (w/w) tannins and not less than 0.6% (v/w) essential oil (*1*).

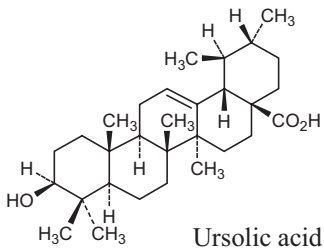
Major chemical constituents
Contains hydrolysable tannins, essential oils, flavonoids, and terpenes (*1*, *6*, *15*). New leaves contain the flavonols quercetin, guajaverin (= quercetin-3-*O*-arabinoside) and other quercetin glycosides; gallocatechin and the tannins ellagic acid and guavins A, C and D. Several triterpene acids are present, including ursolic and oleanolic acids and their 20-hydroxy-derivatives, crataegoic and guaijavolic acids. The leaf oil contains several mono- and sesquiterpenes, among which 1,8-cineol and α-pinene are the

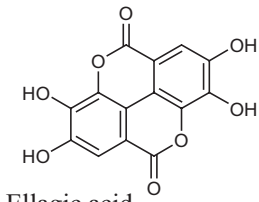
principal monoterpenes, and caryophyllene and γ-bisbolene are representative of the sesquiterpenes (*16–24*).

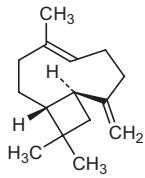
Structures of quercetin, guajaverin (= quercetin-3-*O*-arabinoside), ursolic oleanolic acids, ellagic acid, α-pinene and caryophyllene are presented below.

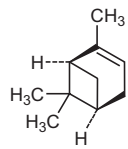



Oleanolic acid


Ursolic acid


Ellagic acid


Caryophyllene



α-Pinene

Medicinal uses

Uses supported by clinical data

Oral treatment of acute diarrhoea, gingivitis and rotaviral enteritis (*25–28*).

Uses described in pharmacopoeias and well established documents

No information was found.

Uses described in traditional medicine

Treatment of abdominal pain, bleeding gums, cough, gastritis, headache, ringworm, vaginitis, wounds and worms. Also used as an astringent, an antiemetic and an emmenagogue (*4*, *15*).

Pharmacology

Experimental pharmacology

Analgesic activity

Intragastric administration of 100.0, 200.0 or 400.0 mg/kg body weight (bw) of the essential oil of the leaves produced an antinociceptive effect in mice as assessed in the formalin test (*29*). A dose of 200.0 mg/kg bw of the essential oil to mice reduced pain as measured in the acetic acid-induced writhing test. One of the major constituents, α-pinene, also showed an antinociceptive effect in the formalin test, and when administered intragastrically at doses of 100 mg/kg and 200 mg/kg, a reduction of paw licking of 72% and 76%, respectively was observed. At a dose of 400.0 mg/kg bw, paw licking was reduced by 37% in the first (acute) phase and 81% in the second (chronic) phase (*29*).

Antidiarrhoeal activity

A decoction of the leaves inhibited Microlax-induced diarrhoea when administered by gastric lavage to rats at a dose of 10.0 ml/kg bw (*30*). Intragastric administration of a methanol extract of the leaves to mice at a dose of 200.0 mg/kg bw prevented castor oil-induced diarrhoea (*31*). Intragastric administration of an aqueous or methanol extract of the leaves to rats at a dose of 400.0 mg/kg bw prevented castor oil-induced diarrhoea, reduced gastric motility and prostaglandin E2-induced enteropooling (*32*). A dried 70% methanol extract of the leaves inhibited the electrically-induced peristaltic reflex of isolated guinea-pig ileum at a concentration of 100 µg/ml (*22*). The main active constituent of the extract was quercetin 1.4×10^{-5} (72.1%), with isoquercetin and hyperin being weakly active (*22*).

Antihyperglycaemic activity

Intragastric administration of a 50% ethanol extract of the leaves to rats, at a dose of 200.0 mg/kg bw, prevented alloxan-induced hyperglycaemia. A butanol fraction of the 50% ethanol extract reduced alloxan-induced hyperglycaemia in rats when administered at a dose of 25.0 mg/kg bw by gastric lavage (*33*). Intragastric administration of a 50% ethanol extract to rats at a dose of 200.0 mg/kg bw did not stimulate insulin biosynthesis (*33*).

Antiinflammatory and antipyretic activity

Intragastric administration of 0.8 ml/kg bw of the leaf essential oil to rats reduced inflammation in the carrageenan-induced oedema of the hind paw test and the cotton pellet granuloma test (*34*). Intragastric administration of a dried methanol extract of the leaves to mice at a dose of 200.0 mg/kg bw reduced carrageenan-induced pedal oedema (*31*). Intra-

gastric administration of a dried methanol extract of the leaves to mice at a dose of 200.0 mg/kg bw reduced yeast-induced pyrexia (*31*).

Antimalarial activity

The following extracts of the crude drug had activity against *Plasmodium falciparum* in vitro with a median inhibitory concentration as follows: ethyl acetate,10.0 µg/ml; 95% ethanol, 36.0 µg/ml; aqueous, 80.0 µg/ml; and petroleum ether, 13.0 µg/ml (*35*, *36*).

Antimicrobial activity

A decoction of the crude drug inhibited the growth of *Carnobacterium gallirarum*, *Escherichia coli*, *Salmonella enteritidis* and *Staphylococcus aureus* with a median inhibitory concentration of 31.25 µg/ml and *Klebsiella pneumoniae*, *Pseudomonas aeruginosa,* and *Shigella flexneri* at a concentration of 62.5 µg/ml (*37*). A tannin fraction separated from an extract of the crude drug had antibacterial activity against *Escherichia coli*, *Citrobacter diversus*, *Klebsiella pneumoniae, Shigella flexneri*, *Salmonella enteritidis* and *Staphylococcus aureus* at concentrations of 60.0–95.0 µg/ml in vitro (*38*). An aqueous extract of the leaves had anti-adhesion activity in *Streptomyces mitis* (70%); these bacteria are involved in the development of dental plaque (*39*).

Antioxidant activity

The antioxidant activity of an aqueous and a 50% ethanol extract of the leaves was assessed using 1,1-diphenyl-2-picrylhydrazyl radical colorimetry. Compared to ascorbic acid, the extracts of the crude drug had a significantly lower antioxidant effect (*40*).

Antispasmodic activity

A dried methanol extract of the leaves reduced spontaneous contractions of isolated rat and guinea-pig ileum when added to the bath media at concentrations of 20.0 ml/l and 80.0 µg/ml, respectively (*30*, *37*, *41*). An aqueous, methanol, or hexane extract of the leaves had smooth muscle relaxant activity in guinea-pig ileum in vitro, at a concentration of 250.0–1000.0 µg/ml (*41*, *42*). A butanol extract of the leaves at a concentration of 0.2 mg/ml reduced acetylcholine-induced contractions in guinea-pig ileum by 95–100% (*43*).

Antitussive activity

Intragastric administration of an aqueous extract of the leaves to rats and guinea-pigs at a dose of 5.0 g/kg bw reduced capsaicin-induced coughing (*44*).

Effects on the central nervous system

Intragastric administration of a dried methanol extract of the leaves to mice at a dose of 200 mg/kg bw reduced acetic acid-induced writhing, thereby demonstrating analgesic activity (*31*). Intragastric administration of a dried methanol extract of the leaves to mice at a dose of 200 mg/kg bw also potentiated phenobarbitone-induced sleeping time (*31*). Intraperitoneal administration of a dried hexane extract of the leaves to mice at a dose of 100 mg/kg bw potentiated sodium pentobarbital-induced hypnosis and increased the latency of leptazol-induced convulsions (*42*). Intragastric or intraperitoneal administration of a dried methanol extract of the leaves at a dose of 3.3 mg/kg bw reduced spontaneous motor activity in mice (*45*). Intragastric administration of dried hexane, ethyl acetate or methanol extracts of the crude drug, at doses of 100.0–1250.0 mg/kg bw produced dose-dependent antinociceptive effects in mice, and prolonged pentobarbitone-induced sleep (*46*).

Haemostatic effects

The effects of an aqueous leaf extract on the bleeding time and the three main mechanisms of haemostasis: vasoconstriction, platelet aggregation and blood coagulation, were investigated. Topical application of the aqueous extract at a concentration of 0.05 µg/ml did not reduce bleeding times in wounded rats. However, the extract (2–6 µg/ml) potentiated the vascular muscle contraction induced by phenylephrine (4.0 µg/ml) in isolated aortic strips from rabbits. The extract also significantly prolonged blood coagulation time in normal plasma treated with 6.0 mg/ml extract in the activated partial thromboplastin time test ($p < 0.05$) (*47*).

Inotropic effects

In guinea-pig atria, an ethanol extract of the leaves reduced atrial contractions by depressing the myocardial force in a concentration-dependent manner (median effective concentration (EC_{50}) = 1.4 g/l). Concentrations higher than 2.5 g/l completely abolished the myocardial contractility. Furthermore, an acetic acid fraction (EC_{50} = 0.07 g/l) of the extract increased the relaxation time measured at 20 and 50% of the force curve by 30 and 15%, respectively, but did not change the contraction time. The negative inotropic effect of the extract was abolished by atropine sulfate, suggesting that either the active substance acts as a cholinergic agonist or that it could release acetylcholine from parasympathetic synapses (*48*).

Toxicity

Intragastric administration of an aqueous extract of the leaves to rats exhibited a median lethal dose of 50.0 g/kg bw (*47*). In chronic toxicity tests,

an aqueous extract of the leaves was administered by gavage to 128 rats of both sexes at doses of 0.2, 2.0 and 20.0 g/day (1, 10 and 100 times the normal therapeutic dose for the treatment of diarrhoea) for 6 months.

The results showed that the body weight gains in male rats were lower in all treated animals. Significant increases in white blood cell count, alkaline phosphatase, serum glutamate pyruvate transaminase and serum blood urea nitrogen levels were observed ($p < 0.05$). Serum sodium and cholesterol levels were significantly reduced ($p < 0.05$) indicating signs of hepatotoxicity. In female rats, serum sodium, potassium and albumin levels increased significantly ($p < 0.05$), while levels of platelets and serum globulin were significantly decreased ($p < 0.05$). Histopathological assessment showed a mild degree of fatty change and hydronephrosis in male rats and nephrocalcinosis and pyelonephritis in female rats (*49*).

Clinical pharmacology

Seventy subjects with gingivitis were enrolled in a 3-week placebo-controlled, double-blind clinical trial to assess the efficacy of a mouthwash containing a decoction of the dried leaves (3 kg in 30 l water boiled for 20 minutes). The placebo mouthwash contained the same ingredients with the exception of the decoction of the leaves. The subjects were stratified into two balanced groups according to their baseline pre-prophylaxis gingivitis scores calculated using the Loe-Silness Gingival Scoring Index. The subjects rinsed their mouth three times daily for 1 minute with 15 ml of their assigned mouthwash. Patients who used the mouthwash containing the leaf extract had less inflammation of the gingiva (19.8%) and fewer sites of severe gingival disease (40.5%) than those using the placebo mouthwash (*26*).

A randomized, controlled clinical trial assessed the efficacy of a decoction of the crude drug for the treatment of infantile rotaviral enteritis. Sixty-two patients with rotaviral enteritis were randomly assigned either to the group treated with the decoction or to the control group. The time until cessation of diarrhoea, the content of sodium in blood, the content of sodium and glucose in stools, and the rate of negative conversion of human rotavirus antigen were recorded. After 3 days, 87% of the subjects in the treated group had recovered, significantly more than the number in the control group (58.1%, $p < 0.05$). The time elapsed until cessation of diarrhoea in the treated group (25.1 ± 9.5 h) was significantly shorter than that for the control group (38.7 ± 15.2 h, $p < 0.01$). The content of sodium and glucose in stools was reduced in the treated group ($p < 0.05$), while the reduction in the control group was insignificant. The rate of negative conversion of human rotavirus in the faeces of the treated group was 87.1%, significantly better than that of the control group (58.1%, $p < 0.05$) (*28*).

An aqueous extract of the crude drug was tested in a clinical study involving small groups of patients aged 5 years and younger or 20–40 years. Patients with acute diarrhoea received the extract, while a comparison group received a kaolin or pectin suspension. The results were similar in all three groups with an efficacy of treatment above 70% (*25*).

A randomized, double-blind, clinical study was performed to evaluate the safety and efficacy of an extract of the crude drug with a standardized content of quercetin. The extract was administered orally to a group of adult patients with acute diarrhoeic disease. Adult patients of both sexes between 20 and 59 years of age suffering from non-complicated acute diarrhoea were included. Acute diarrhoeic disease was defined as a clinical condition characterized by the passing of at least three liquid stools during the previous 24 h and abdominal pain or cramps. Pregnant women and patients with systemic diseases concomitant to acute diarrhoeic disease (such as immunodeficiency and intestinal syndrome), were excluded. The capsules containing 500 mg of the extract were administered every 8 h for 3 days to the treatment group (n = 50), while the control group (n = 50) received capsules of the same size, taste and colour, containing 500 mg of placebo every 8 h for 3 days. Oral rehydration therapy was administered to all patients according to conventional procedures followed in the medical institution for treatment of acute diarrhoeic disease. The results showed that the guava product decreased the duration of abdominal pain in these patients (*27*).

A randomized, double-blind, clinical trial involving 122 patients (64 men and 58 women) was conducted to compare the efficacy of the powdered crude drug with that of tetracycline for the treatment of acute diarrhoea (*50*). The patients were treated with 500 mg of the powdered crude drug (2 capsules of 250 mg each) or matching tetracycline capsules (2 capsules of 250 mg each) every 6 hours for 3 days. The results of the study demonstrated that the powdered drug decreased the stool output, fluid intake and the duration of diarrhoea. The differences between the results from the group treated with tetracycline and the group that received the powdered crude drug were not statistically significant (*50*).

Adverse reactions

One report of allergic dermatitis has been recorded after external application of a tea prepared from the crude drug (*51*).

Contraindications

Hypersensitivity or allergy to the plant material.

Warnings
Do not exceed the recommended dose or duration of treatment (*49*).

Precautions
Carcinogenesis, mutagenesis, impairment of fertility
No information was found.

Pregnancy: teratogenic effects
No information was found.

Pregnancy: non-teratogenic effects
Due to the lack of safety data, the use of the crude drug during pregnancy is not recommended.

Nursing mothers
Due to the lack of safety data, the use of the crude drug during breastfeeding is not recommended.

Paediatric use
Due to the lack of safety data, the use of the crude drug in children aged under 12 years is not recommended.

Other precautions
No information was found.

Dosage forms
Crude drug, decoctions, extracts and teas.

Posology
(Unless otherwise indicated)
As a mouthwash: 15 ml of aqueous extract three times daily for at least 1 minute per session (*26*).

For diarrhoea: 500 mg of the powdered leaf three or four times daily (*50*).

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Lichen Islandicus

Definition
Lichen Islandicus consists of the whole or cut dried thalli of *Cetraria islandica* (L.) Acharius s.l. (Parmeliaceae) (*1*).

Synonyms
Physcia islandica DC, *Lichene islandicus* L. (*2*).

Selected vernacular names
Al kharaza, Blätterflechte, brodmose, cetraria, broedmasa, erba rissa, Fieberflechte, Fiebermoos, focus, hazaz, Heideflechte, Iceland lichen, Iceland liver wort, Iceland moss, Icelandic moss, Isländische Flechte, Isländische Tartschenflechte, Isländisches Moos, kharaz assoukhour, lichen catharticus, lichen d'Islande, lichène islandico, líquén de islandia, Lungenmoos, matmasa, muscus, Purgiermoos, svinmasa (*2–5*).

Geographical distribution
Grows in northern, eastern and central Europe, Siberia and North America (*5*, *6*).

Description
A lichen of approximately 10 cm in height, growing on the ground, the brown shrubby thallus lobed and forked, and with a fringed margin. Upper surface olive-green to brown, with occasional dark reddish brown copular apothecia, and the lower surface whitish-grey with numerous small, whitish depressed spots (*5*, *7*).

Plant material of interest: dried thalli
General appearance
Pieces of foliaceous lichen up to about 15 cm long, composed of numerous erect branches about 6 mm broad or more, in an unevenly developed dichotomy, consists of glabrous, groove-shaped or almost flat, stiff, brittle

bands, 0.3–1.5 cm wide and 0.5 mm thick, sometimes serrated with the margin appearing ciliated (pycnidial) thick; lower surface pale greyish, with scattered small, white, ovoid, depressed spots; occasional, dark reddish-brown, cup-shaped, fruiting bodies (apothecia), about 6 mm in diameter, on the upper surface (greenish to greenish brown) near the margins; texture harsh, springy and brittle; on the apex of the terminal lobes, very rarely, there are brown, discoid apothecia (*1*).

Organoleptic properties

Odour: none; taste: mucilaginous and distinctly bitter (*1*).

Microscopic characteristics

Cut transversely, branches show upper and lower colourless cortical regions composed of closely-packed hyphae, appearing as small-celled pseudoparenchyma; below the upper cortex, the algal or gonidial layer containing numerous yellowish-green, subspherical cells of the alga *Chlorococcum humicola*; a central medulla with more closely-packed, greyish-brown hyphae filaments. Section through an apothecium shows a hymenium layer in the upper cortex with flask-shaped asci, each containing eight ascospores, separated by numerous, narrow, thread-like paraphyses (*3*).

Powdered plant material

A greyish brown powder, abundant fragments of pseudoparenchyma consisting of narrow-lumened, thick-walled hyphae from the marginal layer and wide-lumened hyphae from the adjacent layer consisting of loosely entwined hyphae, in the medullary zone of which, yellowish green algal cells about 15 µm in diameter are embedded; occasionally marginal fragments of the thallus with tube-like or cylindrical spermogonia, up to about 160 µm wide and 400 µm long. Many of the particles stain blue-black with iodine water (*1*).

General identity tests

Macroscopic and microscopic examinations and microchemical tests (*1*, *3*), thin-layer chromatography for characteristic lichen acid profile (*1*), and high-performance liquid chromatographic analysis of protolichesterinic and fumarprotocetraric acids (*8*).

Purity tests

Microbiological

Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*9*).

Foreign organic matter
Not more than 5% (*3*).

Total ash
Not more than 3% (*3*).

Acid-insoluble ash
Not more than 1.5% (*3*).

Water-soluble extractive
To be established in accordance with national requirements.

Loss on drying
Not more than 12% (*10*).

Swelling value
Not less than 4.5 determined on the powdered drug (*1*).

Pesticide residues
The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*1*). For other pesticides, see the *European pharmacopoeia* (*1*) and the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*9*) and pesticide residues (*11*).

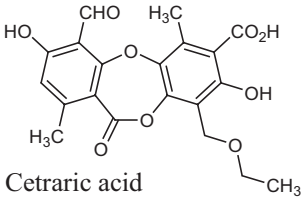
Heavy metals
For maximum limits and analysis of heavy metals, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*9*).

Radioactive residues
Where applicable, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*9*).

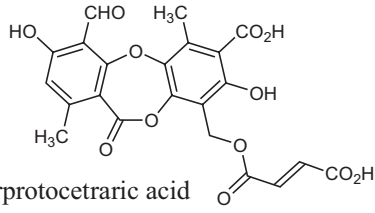
Chemical assays
To be established in accordance with national requirements.

Major chemical constituents
Contains water-soluble polysaccharides (50%), primarily lichenin, a linear cellulose-like polymer of β-D-glucose units, and isolichenin, an α-D-glucose polymer. Two glucans, lichenan and isolichenan, have been isolated from the lichen *Cetraria islandica* (*12*), as well as galactomannans (*5*). Major secondary metabolites include the lichen acids (depsidones),

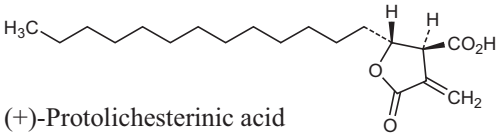
fumarprotocetraric acid, protolichestearic acid and cetraric acids (*5*, *13*). A biologically active aliphatic δ-lactone, (+)-protolichesterinic acid has also been reported (*14*). The structures of fumarprotocetraric acid, cetraric acid and (+)-protolichesterinic acid are presented below.



Cetraric acid



Fumarprotocetraric acid



(+)-Protolichesterinic acid

Medicinal uses

Uses supported by clinical data

Used orally as a demulcent to treat inflammation and dryness of the pharyngeal mucosa (*15*, *16*).

Uses described in pharmacopoeias and well established documents

Used orally for treatment of loss of appetite and nausea (*3*, *17*).

Uses described in traditional medicine

Used to treat asthma, cramps, bronchitis, cough, diabetes, exhaustion, gastric disturbances, immune depletion, migraine, nausea in pregnancy and wounds. Also used as an emergency food source, an emollient and a galactagogue (*4*, *18–20*).

Pharmacology

Experimental pharmacology

Antimicrobial and antiviral activities

An aqueous extract of the thalli and protolichesterinic acid were screened for in vitro activity against *Mycobacterium aurum*, a nonpathogenic organism with a similar sensitivity profile to *M. tuberculosis*. Protolichesterinic acid had a minimum inhibitory concentration (MIC) of ≥ 125.0 µg/ml (*21*). An aqueous extract of the thalli was screened for in vitro activity against *Helicobacter pylori*. (+)-Protolichesterinic acid was identified as

an active component. The MIC range of protolichesterinic acid, in free as well as salt form, was 16.0–64.0 µg/ml (*14*).

Protolichesterinic acid, isolated from the thalli, inhibited the DNA polymerase activity of human immunodeficiency virus-1 reverse transcriptase, with a median inhibitory concentration (IC_{50}) of 24.0 µM (*22*).

Antioxidant activity

The antioxidant activity, reducing power, superoxide anion radical scavenging and free radical scavenging activities of an aqueous extract of the thalli were investigated in vitro. The antioxidant activity increased with increasing amount of extract (from 50.0 to 500.0 µg) added to linoleic acid emulsion. About 50.0, 100.0, 250.0 and 500.0 µg of aqueous extract of the thalli exhibited higher antioxidant activity than 500.0 µg of α-tocopherol. The samples showed 96, 99, 100 and 100% inhibition of peroxidation of linoleic acid, respectively. On the other hand, 500.0 µg of α-tocopherol showed 77% inhibition of peroxidation of linoleic acid emulsion (*23*).

Antiproliferative effects

The effects of protolichesterinic acid, isolated from the thalli, on three malignant cell-lines (T-47D and ZR-75-1 from breast carcinomas and K-562 from erythroleukaemia), as well as normal skin fibroblasts and peripheral blood lymphocytes were assessed in vitro. Protolichesterinic acid caused a significant reduction in DNA synthesis, as measured by thymidine uptake, in all three malignant cell lines. The IC_{50} was between 1.1 and 24.6 µg/ml for protolichesterinic acid. The breast cancer cell-lines were more sensitive than K-562. The proliferative response of mitogen-stimulated lymphocytes was inhibited with an IC_{50} of 24.5 µg/ml for protolichesterinic acid. These concentrations are of the same order of magnitude as the MICs in the 5-lipoxygenase assay. Significant cell death (assessed by the 3-(4,5-dimethylthiazol-2-yl)-5-(3-carboxymethoxyphenyl)-2-(4-sulfophenyl)-2H-tetrazolium assay and trypan blue exclusion) occurred in the three malignant cell-lines at concentrations of protolichesterinic acid greater than 30.0 µg/ml. In K-562, morphological changes consistent with apoptosis were detected. At a concentration of protolichesterinic acid of 20 µg/ml, up to 38% cell death was observed in mitogen-stimulated lymphocytes, but unstimulated lymphocytes were clearly less sensitive. In contrast, the DNA synthesis, proliferation and survival of normal skin fibroblasts were unaffected at doses of protolichesterinic acid of up to 20 µg/ml (*24*).

Immune effects

A polysaccharide, isolated from the lichen, and described as a branched galactomannan, with a backbone composed of two structural elements;

(1→6)-linked α-D-mannopyranosyl and α-D-(1→6)-galactopyranosyl units, had pronounced immunostimulating activity. The polysaccharide enhanced granulocytic phagocytosis by 68%, when administered at a concentration of 1 mg/ml, in human granulocytes in vitro. Intraperitoneal administration of the polysaccharide to mice, at a dose of 1.0 mg/kg body weight (bw), increased the rate of carbon clearance, indicating significant activation of the reticuloendothelial system (*13*, *25*, *26*). A polysaccharide named Ci-3, isolated from an aqueous extract of the thalli, as well as from an alkali extract of the thalli had immune stimulating activity in vitro. At a concentration of 100.0 µg/ml, the polysaccharide Ci-3 stimulated granulocytic phagocytosis and reduced the complement-induced haemolysis in sheep erythrocytes by 80% (*27*).

Toxicity

Lichens are known to naturally contain bitter and potentially toxic lichen acids, as well as concentrated heavy metals that are taken up from the environment. The toxicity of the thalli was investigated using traditional preparation methods (boiling, ash-soaking). Untreated and briefly boiled thalli were lethal to mice when added to feed at concentrations of 25 and 50% w/w, but when the thalli were prepared by ash-soaking, the mice tolerated the crude drug reasonably well for 3 weeks (*28*).

In another study, the toxicity of the traditionally prepared thalli was assessed in mice and rats. As a 50% w/w mixture in normal feed neither the pretreated nor the untreated thalli additive was well tolerated by mice. However, rats tolerated 25% w/w ash-treated thalli in normal feed over 3 months, although the body weight did not increase as much as in control animals. At the end of the experiment, the rats in the thalli-treated group had proteinuria, and on autopsy some tubular changes were found, probably due to high concentrations of lead in the thalli and in the kidneys (*29*).

Clinical pharmacology

In an open clinical trial, 100 patients (children and adults) suffering from inflammatory irritative conditions of the respiratory tract were treated with lozenges containing an aqueous extract of the thalli. The illnesses ranged from pharyngitis and laryngitis to chronic or acute bronchitis. The patients were treated with lozenges containing 160.0 mg of an aqueous extract of the crude drug, for between 4 days and 3 weeks. After evaluation of the symptoms such as cough, hoarseness, secretion, inflammation and pain, a positive outcome was observed in 86% of patients. The lozenges were well tolerated, with few side-effects and good gastric tolerance (*16*).

Sixty-one patients who had recently undergone surgery to the nasal septum were included in a randomized, placebo-controlled, clinical trial.

These patients suffer especially from dryness and inflammation of the oral cavity due to breathing only through the mouth while the nose is permanently closed by a nasal package. Three groups of patients treated with three different concentrations of an aqueous extract of the thalli (0.048 g drug per pastille/day, 0.3 g drug per pastille/day or 0.5 g drug per pastille/day) were compared. Coating, dryness and inflammation of the mucosa, lymph nodes and tongue, the patients' tolerance of the drug, and symptoms such as hoarseness and sore throat were documented. The oral application was performed from the first to the fifth day after the operation. The treatment with the lichen caused a direct reduction of the observed pathological changes. There were no significant differences between the three test groups. A dose of 0.48 g per day (10 pastilles per day) is a sufficient treatment (*15*).

Adverse reactions

Allergic reactions, abdominal pain and nausea have been reported. Few adverse events were observed in a study of 3143 children aged 4–12 years who were administered 320–600 mg of an aqueous extract of the thalli in lozenges per day over a 1–2 week period. The adverse events were mild and transient (*30*).

Contraindications

Hypersensitivity or allergic reactions to the crude drug.

Warnings

No information was found.

Precautions

Carcinogenesis, mutagenesis, impairment of fertility

No information was found.

Other precautions

No information was found.

Dosage forms

Crude drug, extracts, fluidextract, infusion and tinctures (*3*, *17*).

Storage

Store in tightly sealed containers away from light and moisture (*5*).

Posology

(Unless otherwise indicated) (*3*, *17*, *18*)

As a demulcent to treat inflammation and dryness of the pharyngeal mucosa: comminuted herb for infusions and other Galenical formulations for internal use. For loss of appetite: comminuted herb, cold macerates for internal use.

Average daily dosage: 4–6 g of herb in divided doses, or equivalent preparations (*17*). Infusion: 1.5 g of crude drug in 150 ml water. Fluidextract 1:1 (g/ml): 4–6 ml. Tincture 1:5 (g/ml): 20–30 ml (*18*).

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Fructus Macrocarponii

Definition

Fructus Macrocarponii consists of the fresh or dried ripe fruit of *Vaccinium macrocarpon* Ait. (Ericaceae) (*1*).

Note: Cranberry liquid preparation is official in the *United States Pharmacopeia* (*2*).

Synonyms

Oxycoccus macrocarpus (Alt.) Pursh., *O. painstris* var. *macrocarpus* (*1*).

Selected vernacular names

Afonya, American cranberry, bear berry, black cranberry, cranberry, Kronsbeere, large cranberry, low cranberry, marsh apple, Moosbeere, Preisselbeere, suureviljaline jõhvikas, trailing swamp cranberry (*1*, *3*, *4*).

Geographical distribution

Native to eastern North America (*1*).

Description

Trailing evergreen shrub up to 15 cm in height; rhizomatous. Stem: slender, glabrous to hairy, rooting at nodes. Leaf: simple, alternate, sub-sessile; blade narrowly elliptic, rarely oblong, (5–) 7–10 (–18) mm long, (2–) 3–4 (–5) mm wide, apex rounded; leathery, upper surface green, lower surface glaucous; margin entire, rarely revolute. Inflorescence: flowers solitary in leaf axils of new shoots; bracts 2, green, 1–2 mm wide; pedicels 2–3 cm, recurved, jointed to the flower. Flower: perfect, radially symmetrical with a nectariferous disc surrounding the style; calyx 4-lobed; corolla white to pink, cup-shaped, 4-lobed with lobes much longer than cup and strongly reflexed at anthesis; stamens 8, filaments with stiff hairs along margins, anthers dehiscent by apical pores; ovary inferior, style 1, stigma capitate. Fruit: berry, globose, 9–20 mm in diameter, glabrous, red to crimson, dark burgundy or almost black, many-seeded. Chromosome number: $n = 12$ (*1*).

Plant material of interest: dried ripe fruit

General appearance

The dried berry is dark red to almost black, 7.0–11.5 mm in width and 10–15 mm in length, with a smooth but deeply wrinkled, slightly lustrous surface. At the fruit apex is the dried, slightly raised nectariferous disc, 1.5–2 mm across, with a shallow depression in its centre inside which is a small protuberance from the remains of the style. At the fruit base is a small scar where the berry was attached to the pedicel. The texture is spongy. In cross-section, the mesocarp and endocarp are a dull red. The mesocarp has large air pockets, and varies in thickness when dried from 0.1–2 mm. In cross-section, the fruit is divided into 4 chambers (carpels), each running the entire length of the fruit. The chambers are separated by dull red, thin, translucent septa and each chamber contains 1–5 seeds. Each seed is narrowly ovoid with an acute apex, 1–2.7 mm long and approximately 1 mm wide, with a lustrous rose to red testa that is longitudinally wrinkled. The endosperm is an opaque white (*1*).

Organoleptic properties

Odour: slightly fruity and sweet; taste: very acidic and tart (*1*).

Microscopic characteristics

Fruit: the exocarp consists of anthocyanin-coloured polygonal cells covered by a thick cuticle. Groups of cells are separated by fairly thick, colourless walls, whereas the walls within the groups are very thin. The mesocarp consists of large, spherical, thin-walled cells in which small bundles of spirally thickened vessels are embedded.

Seed (cross-section): the epidermis of the testa is composed of radially elongated rectangular cells filled with mucilage. The walls are thickened in a U-shape, the thickest wall being the exterior one; the mucilage is radially striated, and in the centre of the cell is a small lumen. Below the epidermis are several layers of polygonal cells with thick, brown, occasionally reticulately thickened walls. These cells are 250–350 µm long and approximately 80 µm broad. The innermost layer of the testa consists of compressed, rectangular cells with sinuous longitudinal walls. The voluminous endosperm is made up of small polygonal and oil-containing cells (*1*).

Powdered plant material

Numerous fragments of the exocarp with colourless cell walls and violet contents; thin-walled parenchyma of the mesocarp; thickened and pitted cells of the testa; oil droplets (*1*).

General identity tests

Macroscopic and microscopic examinations and high-performance thin-layer chromatography (*1*).

Purity tests

Microbiological

Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*5*).

Foreign organic matter

To be established in accordance with national requirements.

Total ash

To be established in accordance with national requirements.

Acid-insoluble ash

To be established in accordance with national requirements.

Water-soluble extractive

To be established in accordance with national requirements.

Alcohol-soluble extractive

To be established in accordance with national requirements.

Loss on drying

To be established in accordance with national requirements.

Pesticide residues

The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*6*). For other pesticides, see the *European pharmacopoeia* (*6*) and the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*5*) and pesticide residues (*7*).

Heavy metals

For maximum limits and analysis of heavy metals, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*5*).

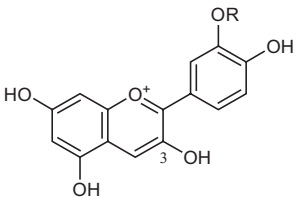
Radioactive residues

Where applicable, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*5*).

Other purity tests
Chemical tests to be established in accordance with national requirements.

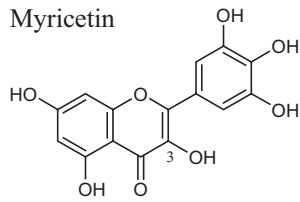
Chemical assays
To be established in accordance with national requirements.

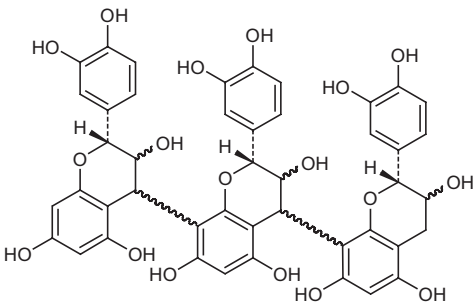
Major chemical constituents
Contains organic acids (e.g. citric and quinic acids predominate), phenolic acids (e.g. chlorogenic and p-coumaric acids). The fruits contain up to 10% tannins (mostly catechol tannins). The major flavonoids are myricetin and quercitin glycoside. The primary constituents of interest in cranberry are the anthocyanins and related compounds based on the anthocyanidins, cyanidin and peonidin, to which are attached a glucose, arabinose or galactose sugar at the number 3 carbon, giving a total of 6 anthocyanins. The galactosides of peonidin and cyanidin predominate, with moderate amounts of the arabinosides. Also of interest are proanthocyanidins (condensed tannins) consisting of oligomers of two flavan-3-ols, primarily epicatechin plus small amounts of epigallocatechin. Cranberry proanthocyanidins contain a variety of complex linkages, including A-type, B-type and branching chains. A-type proanthocyanidins have doubly-linked flavan-3-ol units with both a carbon–oxygen–carbon (C2–O–C7) bond and a carbon–carbon (C4–C8 or C4–C6) bond (*1*, *3*). Structures of cyanidin, peonidin, myricetin and proanthocyanidin trimer are presented below.



Cyanidin
R = H

Peonidin
R = CH_3





Proanthocyanidin (trimer)

Medicinal uses

Uses supported by clinical data

Orally as adjunct therapy for the prevention and symptomatic treatment of urinary tract infections in adults (*8–20*). Two clinical trials have assessed the effect of the fruit juice in paediatric populations (*17*, *21*), but the results were negative. Results from clinical trials involving the use of cranberry for the treatment of children with neurogenic bladder were also negative and do not support the use of cranberry products in paediatric populations (*22*).

Uses described in pharmacopoeias and well established documents

No information was found.

Uses described in traditional medicine

Treatment of asthma, fever, loss of appetite, scurvy and stomach ailments, as well as gallbladder and liver disease and for treatment of wounds (*3*, *23*).

Pharmacology

Experimental pharmacology

Antimicrobial and antiadhesive activity

A concentrated extract of the juice of the crude drug was tested for activity against the following pathogens: *Alcaligenes faecalis*, *Clostridium perfringens*, *Enterococcus faecalis*, *Escherichia coli*, *Mycobacterium phlei, Pseudomonas aeruginosa*, *Salmonella california, Salmonella enteritidis*, *Salmonella typhimurium*, *Shigella sonnei*, *Staphylococcus aureus* and methicillin-resistant *S. aureus*, as well as *Candida albicans.* The crude drug concentrate did not inhibit the growth of *C. albicans*, but had variable activity against the bacteria at dilutions of 1:5 and 1:10 (*24*).

Early investigations of the mechanism of cranberry's bacteriostatic activity suggested that acidification of the urine may be responsible for its activity. However, more recent studies have demonstrated that urinary acidification is not the mechanism by which cranberry exerts its effects, but it does so through a mechanism that involves the inhibition of bacterial adherence (*25–27*). In vitro studies have shown that, in the 77 clinical isolates tested, cranberry juice decreased bacterial adherence of *E. coli* to uroepithelial cells by 60% when compared with saline solution (*28*). Furthermore, cranberry juice inhibited the adherence of *E. coli* to human urinary epithelial cells three times more stongly than *E. coli* isolated from other clinical sources (*27*, *28*). One study demonstrated that cranberry juice dose-dependently inhibited the haemagglutination activity of *E. coli* urinary isolates expressing type I and P adhesion (*27*).

The effect of the fruit on the expression of P-fimbriae of *E. coli* was assessed by growing P-fimbriated *E. coli* in solid media containing cranberry juice. Cranberry concentrate at pH 7.0 was added to the medium to achieve a final concentration of 25%. *E. coli* strains JR1 and DS17 were plated on to this medium, with a plain medium control, and incubated at 37 °C. Cultures were tested for ability to agglutinate P-receptor-specific beads. The results demonstrated that for *E. coli* strain JR1, P-fimbrial agglutination was inhibited after the third plating and strain DS17 was fully inhibited after the second plating. Fully inhibited bacteria had a 100% reduction in expression of fimbriae (*25*). Two compounds in cranberry juice inhibited lectin-mediated adherence of *E. coli* to mucosal cells. One of these compounds was fructose and the other was a nondialysable polymeric compound (*25*). Further investigations have found that exposure of pathogenic bacteria to the nondialysable polymeric compound in either the gut or bladder produces a bacteriostatic effect by inhibiting the expression of specific adhesins present on the pili on the bacterial surface (*26*, *28*).

The proanthocyanidin fraction, isolated from an ethyl acetate extract of the fruit, was assessed for its ability to prevent adherence of *E. coli* by measuring the ability to prevent agglutination of both isolated P-receptor resin-coated beads and human erythrocytes. The proanthocyanidin fraction, at a concentration as low as 75.0 mg/ml, exhibited potent antiadherence activity in uropathogenic isolates of P-fimbriated *E. coli* bacteria to cellular surfaces containing alpha-Gal(1→4)beta-Gal receptor sequences similar to those on epithelial cells in the urinary tract. The chemical structures of the proanthocyanidins consisted predominantly of epicatechin units containing at least one A-type linkage. The procyanidin A2 was the most common terminating unit, occurring about four times as frequently as the epicatechin monomer (*29*). Isolated A-type proanthocyanidins isolated from cranberry juice cocktail had anti-adhesive activity at 60 µg/ml, while the B-type exhibited weak activity at 1.2 mg/ml (*30*).

A high-molecular-weight constituent of the fruit, named nondialysable material, at concentrations of 100.0 µg/ml, inhibited the adhesion of *Helicobacter pylori* strain BZMC-25 to human gastric mucosal cells. The inhibition of adhesion by nondialysable material was dose-dependent, and the 50% inhibitory concentration was strain-dependent, the concentrations being 37.0, 125.0 and 305.0 µg/ml for *H. pylori* strains BZMC-25, EHL-65 and 17874, respectively (*31*).

In one study, uropathogenic P-fimbriated *E. coli* isolates were obtained from the urine of women with clinically diagnosed, culture-confirmed urinary tract infections and incubated for 20 minutes in urine collected over a 12-hour period from healthy women before and after consumption

of 240 ml of commercial cranberry juice cocktail (30% pure juice), and in cranberry proanthocyanidin extract (pH 6.5) (2-fold dilution series). These bacteria were then harvested and screened for ability to adhere to isolated uroepithelial cells and to agglutinate human red blood cells (A1, Rh+), and resin beads coated with isolated P-receptor oligosaccharides. Urine collected from healthy women after consumption of the cranberry juice cocktail (average pH 6.2) prevented adhesion in 31 (80%) of the 39 isolates and 19 (79%) of the 24 antibiotic-resistant isolates in all bioassays, while urine collected prior to the administration of the juice (average pH 6.2) failed to prevent adhesion in any of the samples. Anti-adhesion activity was evident in the urine within 2 hours and persisted for up to 10 hours following ingestion of the cranberry juice cocktail. The extracted proanthocyanidins inhibited adhesion of all isolates at concentrations ranging from 6 to 375.0 µg/ml, demonstrating potent in vitro anti-adhesion activity against these bacterial strains (*32*).

In another study, urine samples were collected from groups of volunteers following the consumption of water, ascorbic acid or cranberry supplements (unspecified) and tested in an anti-adhesion assay. Only intake of ascorbic acid consistently produced acidic urine. Surface tension measurements of the urine collected showed that both water and cranberry supplementation consistently produced urine with a surface tension higher than that in urine from the control group or urine collected following ascorbic acid intake. Urine obtained after supplementation with ascorbic acid or cranberry reduced the initial in vitro deposition rates and numbers of adherent *E. coli* and *Enterococcus faecalis*, but not *Pseudomonas aeruginosa*, *Staphylococcus epidermidis*, or *Candida albicans*. Conversely, urine obtained from subjects with increased water intake vastly increased the initial deposition rates and numbers of adherent *E. coli* and *E. faecalis* ($p < 0.05$) (*33*).

Antioxidant activity

The antioxidant activities of the fruit and its phenolic constituents were measured in vitro. The fruit had an anthocyanin concentration of 0.32 mg/g fresh weight and a total phenolic concentration of 3.15 mg/g fresh weight. In vitro the fruit exhibited antioxidant effects at a concentration of 18.5 µmol/g fresh weight. Chlorogenic acid, peonidin 3-galactoside, cyanidin 3-galactoside and cyanidin 3-galactoside were the most important antioxidants in the fruit (*34*). Methanol extracts of the fruit were assayed for radical-scavenging activity and cell growth inhibition using seven tumour cell lines. A methanol extract of the fruit caused inhibition of the proliferation of K562 and HT-29 cell lines at concentrations in the range of 16.0–125.0 µg/ml. Radical-scavenging activity was greatest in an extract composed primarily of flavonol glycosides. Seven flavonol mono-

glycosides: myricetin 3-alpha-arabinofuranoside, quercetin 3-xyloside, 3-methoxyquercetin 3-beta-galactoside (isorhamnetin), myricetin 3-beta-galactoside, quercetin 3-beta-galactoside, quercetin 3-alpha-arabinofuranoside and quercetin 3-alpha-rhamnopyranoside were evaluated for 1,1-diphenyl-2-picrylhydrazyl radical-scavenging activity and ability to inhibit low-density lipoprotein oxidation in vitro. Most of the flavonol glycosides had antioxidant activity comparable to or superior to that of vitamin E. Cyanidin 3-galactoside showed activity superior to that of the flavonoids as well as vitamin E or of a water-soluble vitamin E analogue in both antioxidant assays (*35*).

In another in vitro study, the antioxidant and anti-inflammatory effects of cranberry anthocyanins and hydroxycinnamic acids were measured against hydrogen peroxide- and tumour necrosis factor (TNFα)-induced damage to human microvascular endothelial cells. Polyphenols from the crude drug were absorbed into endothelial cells and subsequently reduced the vulnerability of endothelial cells to increased oxidative stress in the membrane and cytosol. Furthermore, the polyphenols also reduced TNFα-induced up-regulation of various inflammatory mediators (interleukin-8) involved in the recruitment of leukocytes to sites of damage or inflammation along the endothelium. Maximum inhibition for all treatments was observed at a concentration of 0.1 mg/ml for 2 hours ($p < 0.001$ in all cases) (*36*).

Clinical pharmacology

Numerous clinical studies have assessed the effects of the fruit and of fruit juice preparations in humans, but only 18 clinical trials have assessed the effects of cranberry juice on urinary pH and urinary tract infections (*8–20*, *37*). Of these 18 trials assessing the safety and efficacy of cranberry for the prevention and treatment of urinary tract infections (UTIs), only four were controlled and of sufficient scientific quality (*8*, *14*, *16*, *19*, *38*).

A double-blind, placebo-controlled trial involving 376 elderly patients in hospital assessed the prophylactic effects of cranberry juice (30% pure juice, 300 ml per day) against the onset of urinary tract infections (*37*). Although the results suggested that cranberry supplementation may be protective, these results were not statistically significant.

A randomized placebo-controlled clinical trial compared the effectiveness and cost-effectiveness of concentrated cranberry tablets (1:30 pure dried juice), cranberry juice, and a placebo used as prophylaxis against lower urinary tract infections in adult women. One hundred and fifty sexually active women aged between 21 and 72 years were randomly assigned for one year to one of three groups receiving prophylaxis: placebo juice + placebo tablets; placebo juice + cranberry tablets; or cranberry

juice + placebo tablets. Tablets were taken twice daily; juice was administered at a dose of 250 ml three times daily. Outcome measures were:

— a > 50% decrease in number of symptomatic UTIs per year (symptoms ≥ 100 000 single organisms/ml); and
— a > 50% decrease in annual consumption of antibiotics.

Cost-effectiveness was calculated as cost in US dollars per urinary tract infection prevented. Both cranberry juice and cranberry tablets led to a statistically significant decrease in the number of patients experiencing at least one symptomatic UTI per year (to 20% and 18%, respectively) compared with placebo (to 32%) ($p < 0.05$) (*19*).

An open, randomized controlled 12-month follow-up trial was performed to determine whether recurrences of urinary tract infection could be prevented with cranberry–lingonberry juice or with a lactobacillus drink. One hundred and fifty women, with UTIs caused by *E. coli,* were allocated to one of three groups. The first group received 50.0 ml of cranberry–lingonberry juice concentrate mixed in 200.0 ml water (7.5 g cranberry concentrate and 1.7 g lingonberry concentrate in 50.0 ml water, no sugar added) daily for 6 months; the second, 100 ml of lactobacillus drink 5 days per week for one year; and the third group received no intervention. Outcomes measured included the first recurrence of symptomatic UTI, defined as bacterial growth of $\geq 10^5$ colony-forming units/ml in a clean voided midstream urine specimen. The cumulative rate of the first recurrence of UTI during the 12-month follow-up period differed significantly between the groups ($p = 0.048$). After 6 months, eight (16%) women in the group treated with cranberry, 19 (39%) in the lactobacillus-treated group, and 18 (36%) in the control group had had at least one recurrence. A 20% reduction in absolute risk in the group given cranberry was observed as compared with the control group (*14*).

A randomized, double-blind, placebo-controlled parallel trial involving 153 elderly female volunteers (mean age 78.5 years) assessed the effect of a cranberry juice preparation (27% pure juice, saccharin-sweetened) on asymptomatic bacteriuria (defined as $> 10^5$ colony forming units/ml) and pyuria (*38*). The subjects were randomly assigned to receive 300 ml/day of cranberry juice or a cranberry-flavoured placebo containing vitamin C for 6 months. Urine samples were collected at monthly intervals. The subjects receiving the cranberry juice had a lower frequency of bacteriuria with pyuria than the subjects in the control group (odds ratio 42% of the control group, $p = 0.004$), of nearly 50% after 4–8 weeks of cranberry use. There was no evidence of urinary acidification, and the median pH of urine in the cranberry-treated group was 6.0.

A randomized, double-blind, placebo-controlled trial assessed the effects of capsules containing concentrated cranberry (400.0 mg of cranberry solids) in 19 women with a history of recurring UTI (*20*). Half of the women were given two capsules of a dried extract of the crude drug daily for 3 months followed by 3 months of treatment with a placebo. The other group received a placebo for the first 3 months then the active treatment for a further 3 months. However nine subjects dropped out due to pregnancy or unrelated infections, or were lost to follow-up. Treatment with cranberry capsules significantly reduced the occurrence of UTIs ($p < 0.005$). On average, 2.4 UTIs per year were recorded in the cranberry-treated group and 6.0 UTIs per year occurred in the placebo-treated group. Treatment was well tolerated and no side-effects were reported.

A pilot study of 15 patients who had spinal cord injuries was performed to determine whether alteration of fluid intake and use of cranberry juice altered the bacterial biofilm load in the bladder. Urine samples were collected on day 0 (start of study), on day 7 – after each patient had drunk one glass of water three times during the day in addition to the normal diet, and on day 15 – after each patient had drunk one glass of cranberry juice three times during the day. The results showed that cranberry juice intake significantly reduced the biofilm load compared to baseline ($p = 0.013$). This was due to a reduction in adhesion of Gram-negative ($p = 0.054$) and Gram-positive ($p = 0.022$) bacteria to cells. Water intake did not significantly reduce the bacterial adhesion or biofilm presence (*16*).

A randomized, placebo-controlled trial assessed the efficacy of 30 ml of pure cranberry juice for reducing the bacterial concentrations in the urine of elderly subjects with a mean age of 81 years (*10*). Thirty-eight volunteers were treated with 30 ml of cranberry juice mixed with water or with water alone for 4 weeks, followed by cross-over for a further 4 weeks. Statistically significant results were reported; cranberry treatment decreased the frequency of bacteriuria ($p = 0.004$). However, 21 patients dropped out before the end of the trial, leaving only 17 patients in the final evaluation. No side-effects were reported (*10*).

In an uncontrolled study involving 60 subjects with symptoms of acute UTI such as frequency, dysuria, urgency and nocturia, the effects of cranberry on the numbers of bacteria in the urine was assessed (*15*). Patients were treated with 16 ounces of cranberry juice daily for 21 days. After 3 weeks, a positive clinical response (no urogenital complaints and fewer than 100 000 bacteria per ml of urine) was noted in 32 patients (53%). Another 12 patients (20%) were "moderately improved" and 16 patients (27%) showed no bacteriological improvement or symptomatic relief (*15*).

In an uncontrolled study, 28 patients in a nursing home were treated with 4–6 ounces of cranberry juice (30% juice) daily for 7 weeks (*9*). Twice-weekly urine samples were examined for leukocytes and/or nitrates as a measure of UTI. At the end of 7 weeks, 10 patients had no leukocytes or nitrates in the urine; nine patients had from a trace to 2+ leukocytes and no nitrates; nine had a trace or greater number of leukocytes. However, this study did not include a non-exposure cohort (control) (*9*).

A number of uncontrolled observational studies have assessed the effects of cranberry juice (33% juice) on urinary pH (*12*, *13*). In the study by Kinney & Blount (*13*), 59 patients (40 of whom completed the study) were treated with 450–720 ml of a preparation containing 80% cranberry juice per day for 6 days, followed by 6 days of no juice, and the pH of their urine was measured. A decrease in urine pH was observed, but it was not dose-related. The second study involved four healthy volunteers who were administered from 1.5 to 4.0 l per day of a 33% cranberry juice product (*12*). Three of the four subjects showed transient changes in urine pH, from 6.6 to 6.33 ($p = 0.01$) and titratable acidity (*12*).

A randomized, double-blind, placebo-controlled study was conducted on 48 patients with neurogenic bladder secondary to spinal cord injury (*39*). Twenty-six of the patients received 2 g of cranberry juice concentrate and 22 received placebo. After 6 months of treatment, bacteriuria and pyuria were not reduced (*39*).

Paediatric populations

An uncontrolled study assessed the efficacy of cranberry (30% pure juice) in 17 children with spina bifida who were using either an indwelling catheter or intermittent self-catheterization (*21*). The children received one, two or three glasses of cranberry juice over a 2-week period. The results of this study showed a reduction of white and red blood cell counts in nearly all urine samples; however the urine from most of the children remained positive for *E. coli* (*21*).

At least two controlled clinical trials have assessed the effects of cranberry in children with neurogenic bladder. A randomized single-blind, cross-over study assessed the efficacy of 15 ml/kg body weight/day of cranberry cocktail juice (30% concentrate) as prophylaxis for bacterial UTIs in 40 children with neuropathic bladder, managed by intermittent catheterization (*8*). The subjects were treated for 6 months with either cranberry juice or water as a control. Outcomes measured were a positive or negative urine culture with symptomatic UTI. The results of this study did not support the use of cranberry juice as prophylaxis against UTIs in children with neuropathic bladder. However, 19 subjects dropped out of

the trial, and the diagnostic criteria for the UTI were much lower than those used in any other trial (10^3 colony-forming units/l of a pathogenic organism).

The results of this trial were confirmed in a double-blind, placebo-controlled, cross-over study involving 15 paediatric patients with neurogenic bladder receiving clean intermittent catheterization (*17*). Two ounces of cranberry juice concentrate (equal to 300 ml of cranberry juice cocktail) or a placebo were administered daily for 3 months, followed by a 3-month cross-over (no washout period was described). Weekly home visits were made, during which a sample of bladder urine was obtained by intermittent catheterization. Signs and symptoms of UTI and all medications were recorded, and compliance was assessed. The results of the study demonstrated that during consumption of the cranberry concentrate, the frequency of bacteriuria remained high. Cultures of 75% (114) of the 151 samples obtained during consumption of placebo were positive for a pathogen ($\geq 10^4$ colony-forming units/ml) compared with 75% (120) of the 160 samples obtained during consumption of cranberry concentrate. *Escherichia coli* remained the most common pathogen during periods of consumption of placebo and cranberry. Three symptomatic infections occurred during the period of treatment with the placebo and three during the period of treatment with the cranberry juice. No significant difference was observed between the acidification of urine in the placebo-treated group and that in the cranberry-treated group (median, 5.5 and 6.0, respectively). Thus, cranberry ingestion did not reduce bacteriuria or symptomatic UTIs in this population (*17*). Unfortunately the only outcomes measured were bacteriuria and presence of symptoms and no blood cell counts were performed.

A review of these clinical data on cranberry juice and supplements in paediatric populations indicates that these products are not effective for the treatment of UTIs in children with neuropathic bladder (*22*).

In an uncontrolled trial involving 13 urostomy patients, the effects of cranberry juice on skin complications were assessed (*40*). Patients were treated with 160 to 320 ml of cranberry juice daily for 6 months. The results of this study showed an improvement of the skin conditions in four urostomy patients with peristomal skin disorders. A decrease in erythema, maceration and pseudoepithelial hyperplasia was observed, but no effects on urine acidity were noted (*40*).

A study was performed to assess the effects of cranberry on the complications of long-term indwelling bladder catheterization (e.g. encrustation and blockage by crystalline *Proteus mirabilis* biofilms). Urine was collected from groups of volunteers who had been treated with up to

1000 ml of cranberry juice or water over an 8-hour period. Laboratory models of the catheterized bladder were supplied with urine from these groups and then inoculated with *P. mirabilis.* After incubation for 24 or 48 hours, the extent of catheter encrustation was determined by chemical analysis for calcium and magnesium. The amount of calcium and magnesium recovered from catheters incubated in urine pooled from individuals who had drunk 500 ml of cranberry juice did not differ significantly from that on catheters incubated in pooled urine from control subjects who had drunk 500 ml of water. However, there was significantly less encrustation ($p = 0.007$) on catheters from models incubated in urine from volunteers who had been given 1000 m1 of water than on catheters incubated in models supplied with urine from volunteers who had been given 1000 ml of cranberry juice. The amounts of encrustation on these two groups of catheters were also significantly less than that on catheters incubated in models supplied with urine from volunteers who had not supplemented their normal fluid intake ($p < 0.001$) (*41*).

Urolithiasis

The potential influence of cranberry juice on urinary biochemical and physicochemical risk factors associated with the formation of calcium oxalate kidney stones was assessed in a randomized cross-over trial. Urinary variables were assessed in 20 South African men with no previous history of kidney stones. The first group of 10 subjects drank 500 ml of cranberry juice diluted with 1500 ml tap water every day for 2 weeks, while the second group drank 2000 ml of tap water daily for the same period. This was followed by a 2-week washout period before the two groups were crossed over. During the experimental phase, subjects kept a 3-day food diary to assess their dietary and fluid intakes; 24-hour urine samples were collected at baseline and on day 14 of the trial periods, and analysed using modern laboratory techniques. Urine analysis data were used to calculate the relative urinary supersaturations of calcium oxalate, uric acid and calcium phosphate. The results demonstrated that the ingestion of cranberry juice decreased oxalate and phosphate excretion and increased citrate excretion. There was also a decrease in the relative supersaturation of calcium oxalate, which tended to be significantly lower than that induced by water alone (*42*).

In another study, five healthy volunteers on a normal diet provided 24-hour urine samples for analysis of pH, volume, creatinine, oxalate, calcium, phosphate, uric acid, sodium, citrate, magnesium and potassium. Tablets containing the crude drug were administered to these volunteers at the manufacturer's recommended dosage (not stated) for 7 days. On the seventh day, a second 24-hour urine sample was obtained from each

volunteer. The results demonstrate that urinary oxalate levels in the volunteers significantly increased ($p = 0.01$) by an average of 43.4% during treatment. The excretion of potential lithogenic ions, calcium, phosphate, and sodium also increased. However, inhibitors of stone formation, magnesium and potassium, rose as well (*43*).

The effect of cranberry juice on risk factors for urinary stones was assessed in a trial involving 12 healthy male subjects. The subjects provided 24-hour urine samples (three samples from each patient during the three loading phases with water only). In each loading phase a neutral mineral water was substituted for 330 ml of the juice. Cranberry juice decreased the urinary pH, whereas the excretion of oxalic acid and the relative supersaturation for uric acid were increased, indicating the potential for the development of urinary stones (*44*).

Adverse reactions

One rare case of immune-related thrombocytopaenia has been reported in a 68-year-old patient (*45*).

Nausea and severe vomiting were reported in a 4-month-old infant after administration of 180 ml of cranberry juice (*46*).

Contraindications

The use of cranberry for the treatment of diseases of the urinary tract in paediatric populations is ineffective and is not recommended (*22*, *43*).

Warnings

Patients with kidney stones or impaired kidney function should use products containing the fruit only after consulting their health care provider (*47*).

Precautions

A health care professional should be consulted prior to treatment to rule out serious conditions such as pyelonephritis.

People with diabetes should be aware of the high content of sugar in the juice and use sugar-free preparations.

Drug interactions

One case of a fatal internal haemorrhage was reported in a male patient whose international normalized ratio (INR) increased to > 50 after being treated with cefalexin, digoxin, phenytoin and warfarin, as well as cranberry juice for 2 weeks. The subject was also unable to eat during the 2-week period (*48*). One case report of a possible interaction with warfa-

rin was reported in a 69-year-old male patient with a mitral valve replacement (*49*). The patient was ingesting 2 l of cranberry juice per day in addition to warfarin and his INR ranged between 8 and 11. The patient was advised to stop drinking cranberry juice and his INR returned to 3 (normal) after 3 days.

The Medicines and Healthcare Products Regulatory Agency (MHPRA) in the United Kingdom has reported numerous cases of a possible interaction between cranberry juice and warfarin, leading to changes in the INR or to bleeding. In four cases, the increase in the INR or bleeding was not significant. Two cases of an unstable INR and one case of a decreased INR were reported (*48*).

Carcinogenesis, mutagenesis, impairment of fertility

No information was found.

Other precautions

No information was found.

Dosage forms

Crude drug, extracts, juice, tablets, capsules. Store in a well-closed container, in a refrigerator (*2*).

Posology

(Unless otherwise indicated)

For the prevention of UTIs in adults the recommended daily dose of cranberry juice is 30–300 ml of a 30% pure juice product; for the treatment of UTIs in adults the daily dosage range is 360–960 ml or equivalent (*1*).

Capsules containing a concentrated cranberry extract: 1–6 capsules daily, equivalent to 3 fluid ounces (90 ml) cranberry juice or 400–450 mg cranberry solids (*1*).

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Cortex Magnoliae

Definition

Cortex Magnoliae consists of the dried stem, trunk or root bark of *Magnolia officinalis* Rehder and Wilson, *M. officinalis* Rehder and Wilson var. *biloba* Rehder and Wilson (*1–3*), or of *M. obovata* Thunberg. (*1*, *3*, *4*) (Magnoliaceae).[1]

Synonyms

Magnolia hypoleuca Diels, non Sieb. et Zucc. (*6*, *7*).

Selected vernacular names

Chung-pu, danghoobak, hou-po, hou-pu, hòupò, hsin-j, koboku, magnolia bark, mubak (*8–11*).

Geographical distribution

Native to China (*7*, *9*, *11*).

Description

Deciduous tree, large, up to 22 m in height. Bark smooth, light rusty-ash grey colour and aromatic. Branchlets light-green or yellowish. Leaves entire, very large, elliptic-obovate, up to 35 cm long and 10–20 cm wide. Large creamy-white, fragrant flowers, bisexual, 15–20 cm in diameter; 9–15 petals, 8–10 cm in length by 3 cm in width, with the outer 3 being pale green tinged pink on the edge and the inner 6–12 being creamy white. Fruit oblong-ovoid, 10–12 cm in length, apex truncate and base rounded. Carpels rounded at the base. Seeds single (*7*, *9*).

[1] The flowers of *Magnolia officinalis* Rehder and Wilson and *M. officinalis* Rehder and Wilson var. *biloba* Rehder and Wilson are also official in the Chinese Pharmacopoeia and the Japanese Standards for Herbal Medicines (*2*, *3*, *5*). However, there is currently insufficient information available to warrant the preparation of a monograph on this material.

Plant material of interest: dried stem, trunk or root bark

General appearance

Trunk bark: rough, 2–7 mm thick, plate-like or semi-tubular bark rolled into large, tight cylinders. The outer surface is greyish white to greyish brown, and rough, sometimes cork layer removed, and externally brown. Inner surface smooth, light brown to purplish brown. Cut surface extremely fibrous, light reddish brown to purplish brown (*1*, *3*, *4*). Stem bark: quilled or double-quilled, 30–35 cm long, 2–7 mm thick, commonly known as *tongpu*. Root bark: quilled singly or irregular pieces some curved like chicken intestine, commonly known as *jichangpo* (*2*).

Organoleptic properties

Odour: aromatic; taste: pungent and slightly bitter (*1–4*).

Microscopic characteristics

Transverse section reveals a thick cork layer or several thin cork layers. The outer surface of the cortex shows a ring of stone cells and scattered on the inner surface are numerous oil cells and groups of stone cells. Phloem rays 1–3 cells wide; fibre groups, mostly several in bundles, scattered. Bast fibre groups lined stepwise between the medullary rays in the secondary cortex. Most of the oil cells are scattered in the primary cortex, with small numbers scattered in the secondary cortex, but some can also be observed in the narrow medullary rays (*1–4*).

Powdered plant material

Yellow-brown powder consisting of a yellowish to red-brown cork layer; stone cells of various sizes or groups; numerous fibres, 12–32 μm in diameter, walls strongly thickened, sometimes undulate or serrate on one side, 1ignified, pit canals indistinct; oil cells containing a yellowish-brown to red-brown substance; single starch grains of about 10 μm in diameter and 2- to 4-compound starch grains, and parenchyma cells containing starch grains (*1*, *2*, *4*).

General identity tests

Macroscopic and microscopic examinations (*1–4*), microchemical test (*4*) and thin-layer chromatography (*1–3*).

Purity tests

Microbiological

Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*12*).

Chemical
To be established in accordance with national requirements.

Foreign organic matter
To be established in accordance with national requirements.

Total ash
Not more than 6% (*1*, *3–5*).

Acid-insoluble ash
To be established in accordance with national requirements.

Water-soluble extractive
To be established in accordance with national requirements.

Alcohol -soluble extractive
Not less than 12% (*1*).

Loss on drying
To be established in accordance with national requirements.

Pesticide residues
The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*13*). For other pesticides, see the *European pharmacopoeia* (*13*), and the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*12*) and pesticide residues (*14*).

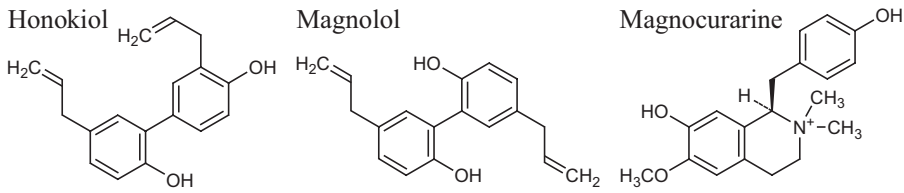
Heavy metals
For maximum limits and analysis of heavy metals, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*12*).

Radioactive residues
Where applicable, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*12*).

Chemical assay
Total magnolol and honokiol not less than 2.0% by high-performance liquid chromatography (*2*).

Major chemical constituents
Magnolol (0.01–16%) and honokiol (0.005–9%) are bioactive major constituents belonging to the lignan group of compounds. Another major

constituent is the isoquinoline alkaloid, magnocurarine (0.15–0.23%). The presence of liriodendrine has also been reported. The bark also contains an essential oil, the major constituents of which are cadinol (14%), 1,4-cineole (6%), *p*-cymene (8%), and β-eudesmol (17%) and geraniol (9%), among others (*8*, *15–18*). The structures of magnolol, honokiol and magnocurarine are presented below.



Medicinal uses

Uses supported by clinical data

No information was found.

Uses described in pharmacopoeias and well established documents

Used orally for the treatment of gastrointestinal disorders such as constipation, dyspepsia, gastritis, nausea and vomiting. Also used orally to treat anxiety, coughs and shortness of breath (*2*).

Uses described in traditional medicine

Treatment of allergic rhinitis, headache, lack of appetite, respiratory congestion, neurosis and fever, and as a uterine stimulant (*6*, *8*, *19*, *20*).

Pharmacology

Experimental pharmacology

Since no clinical studies have directly evaluated Cortex Magnoliae for any therapeutic condition and very few pharmacological studies have been conducted on extracts of the bark, most of this section describes the pharmacology and clinical studies of the major chemical constituents, particularly magnolol. The correlation of these data to the crude drug or its extracts requires further investigation.

Anti-allergic activity

Oral administration of 0.01–1.0 g/kg body weight (bw) of an aqueous extract of the bark dose-dependently inhibited compound 48/80-induced systemic anaphylaxis in rats (*21*). At the same dose, the aqueous extract

also significantly inhibited local immunoglobulin E (IgE)-mediated passive cutaneous anaphylactic reaction and reduced the levels of plasma histamine in a dose-dependent manner ($p < 0.05$). In vitro, the extract (at concentrations of 0.001–1.0 mg/ml) concentration-dependently inhibited the histamine release from rat peritoneal mast cells activated by compound 48/80 or anti-dinitrophenyl IgE (*21*).

Anti-asthmatic activity

Magnolol stimulates calcium channel activity in tracheal smooth muscle cells as assessed by the patch clamp technique (*22*). In whole-cell current recordings, magnolol reversibly increased the amplitude of potassium outward currents. The increase in outward current caused by magnolol was sensitive to inhibition by iberiotoxin (200 nM) or paxilline (1 μM), but not by glibenclamide (10 μM). In inside-out patches, addition of magnolol to the bath did not modify single channel conductance, but effectively enhanced the activity of large conductance calcium (Ca) activated potassium BK (Ca) channels. Magnolol increased the probability of these channel openings in a concentration-dependent manner with a median effective concentration (EC_{50}) value of 1.5 μM. The direct stimulation of these BK (Ca) channels by magnolol may explain the mechanism by which it acts as an anti-asthmatic compound (*22*).

Antibacterial activity

An ethanol extract of the bark inhibited the growth of *Actinomyces viscosus* ATCC 19246, *Streptococcus mutans* Ingbritt and *Streptococcus sobrinus* 6715, with a minimum bactericidal concentration of 0.488, 0.488 and 1.250 g/l, respectively (*23*). The antimicrobial activities of honokiol and magnolol were assayed using the agar dilution method and the minimum inhibitory concentration (MIC) was determined for each compound using a twofold serial dilution assay. The results showed that honokiol and magnolol have a marked antimicrobial effect (MIC, 25.0 μg/ml) against *Actinobacillus actinomycetemcomitans*, *Porphyromonas gingivalis*, *Prevotella intermedia*, *Micrococcus luteus* and *Bacillus subtilis*, but were not active against *Shigella flexneii*, *Staphylococcus epidermidis*, *Enterobacter aerogenes*, *Proteus vulgaris*, *Escherichia coli* and *Pseudomonas aeruginosa* (*24*). An extract of the bark and magnolol inhibited the growth of *Helicobacter pylori* in vitro (*25*).

Anti-gastric ulcer activity

Intragastric administration of aqueous and methanol extract of the crude drug (400 mg/kg bw) reduced gastric juice secretion and increased the pH of gastric secretions in mice pretreated with indometacin (*26*). Indometa-

cin-induced gastric ulceration was reduced in animals treated with 400 mg/kg bw of the methanol extract (*26*). Intragastric administration of an ethanol extract of the crude drug at a dose of 5.0 or 15.0 g/kg bw inhibited hydrochloric acid-induced gastric ulceration in mice (*27*).

Anti-inflammatory activity

Magnolol, isolated and purified from the crude drug, inhibited mouse hind-paw oedema induced by carrageenan, compound 48/80 and polymyxin B and reversed passive Arthus reaction when administered orally at a dose of 30 mg/kg bw (*28*).

Antioxidant activity

The accumulation of oxygen-free radicals and activation of neutrophils are implicated in the pathophysiological mechanisms that mediate myocardial ischaemia/reperfusion injury. Thus, antioxidants are purported to have cardioprotective activity. The antioxidant effect of magnolol was evaluated in an open-chest anaesthetized rat model of myocardial ischaemia/reperfusion injury (*29*). Intravenous pretreatment with magnolol, at a dose of 0.2 and 0.5 µg/kg bw at 10 minutes prior to 45 minutes of left coronary artery occlusion, reduced the incidence and duration of ventricular fibrillation and reduced mortality when compared with the control group. After 1 hour of reperfusion, pretreatment with magnolol reduced infarct size. In addition, magnolol, at a dose of 0.2 µg/kg bw, reduced superoxide anion production and myeloperoxidase activity, an index of neutrophil infiltration in the ischaemic myocardium (*29*).

Restenosis, a common complication after balloon angioplasty, involves a number of cytokines, chemotactic factors and growth factors. Antioxidants have been shown to inhibit intimal thickening after balloon injury in hyperlipidaemic animals. The effects of magnolol on the expression of monocyte chemotactic protein-1 and on intimal response in balloon-injured aorta of cholesterol-fed rabbits were investigated. The animals were fed a 2% high-cholesterol diet together with daily intramuscular injection of either 1 µg/kg bw of magnolol or vehicle solvent for a total of 6 weeks, while 10 rabbits fed a regular diet served as a control group. A balloon denudation of abdominal aorta was performed in each group at the end of the third week, and aortas were harvested at the end of 6 weeks. Treatment with magnolol significantly inhibited copper-induced low-density lipoprotein oxidation in cholesterol-fed rabbits and reduced atheroma formation ($p < 0.05$) in thoracic aortas without lowering serum cholesterol. The intimal response was significantly attenuated in magnolol-treated rabbits receiving high cholesterol when compared to those of the control high-cholesterol group ($p < 0.05$) (*30*).

The protective effect of magnolol against hypoxia-induced cell injury in cortical neuron-astrocyte mixed cultures was assessed after exposure of the cells to chemical hypoxia (0.5 mm potassium cyanide). Treatment with magnolol (10 and 100 μM) significantly reduced potassium cyanide-induced lactose dehydrogenase release in a concentration-dependent manner (*30*).

The effects of magnolol on the course of sepsis, survival rate and biochemical parameters were analysed in rats with induced sepsis. Intragastric administration of magnolol, at doses ranging from 10^{-9} g/kg to 10^{-5} g/kg bw, administered either before or after induction of sepsis by cecal ligation and puncture did not alter the course of sepsis induced by two cecal punctures. However, when one cecal puncture was performed, a moderately evolving type of sepsis was induced, and the survival rate of affected rats was significantly improved by pretreatment with 10^{-7} g/kg magnolol ($p < 0.05$). The intensity of lipid peroxidation in plasma, liver and lung of septic rats was also attenuated in a treatment-dependent manner (*31*).

Anxiolytic activity

In rat brain membranes, honokiol and magnolol at a concentration of 5.0 μM, enhanced binding of [3H]muscimol and [3H]flunitrazepam to the γ-aminobutyric acid receptor, but honokiol was 2.5 to 5.2 times more potent than magnolol. Honokiol and magnolol also enhanced the potentiating effect of 200 nM γ-aminobutyric acid on [3H]flunitrazepam binding with EC_{50} values of 0.61 μM and 1.6 μM. Honokiol and magnolol increased [3H]muscimol binding by approximately 68% with EC_{50} values of 2.3 and 12.0 μM, respectively (*32*).

In elevated plus-maze tests, treatment of mice with honokiol prolonged the time spent in the open arms, showing an anxiolytic effect. Seven daily treatments with 0.1–1 mg/kg bw honokiol, demonstrated an anxiolytic effect with the optimal dose at 0.2 mg/kg bw. These results suggest that honokiol is the chemical responsible for the anxiolytic effect of the water extract of the bark in vivo (*33*).

Cardiovascular effects

The effect of magnolol on coronary circulation and vascular resistance was assessed in anaesthetized rabbits using a Doppler velocimetry probe. Thirty-nine rabbits received intravenous injection of either vehicle ($n = 5$), magnolol (10^{-6}–10^{-4} g/kg bw) or nitroglycerin. Magnolol did not change blood pressure or coronary blood flow velocity, but at a dose of 10^{-4} g/kg, decreased coronary vascular resistance significantly more than vehicle ($p < 0.001$). Nitroglycerin increased coronary blood flow velocity and

decreased coronary vascular resistance in a dose-dependent manner ($p < 0.01$) (*34*). Magnolol (concentration 10–100 µg/ml) inhibited norepinephrine-induced phasic and tonic contractions in rat thoracic aorta in vitro (*35*).

Miscellaneous activities

Honokiol and magnolol increase choline acetyl-transferase activity, inhibit acetylcholinesterase, promote potassium-induced acetylcholine release and exhibit neurotrophic function in vitro. In conscious, freely-moving rats a dose of honokiol or magnolol (10^{-4}–10^{-6} M) was perfused into the hippocampus via a dialysis probe. The results showed that at a dose of 10^{-4} M honokiol or magnolol markedly increased extracellular acetylcholine release to 165.5% and 237.83% of the basal level, respectively. However, lower concentrations of either compound failed to elicit significant acetylcholine release (*36*).

The effects of magnolol on thermoregulation and hypothalamic release of 5-hydroxytryptamine were assessed in normothermic rats and in febrile rats treated with interleukin-1 beta. Intraperitoneal administration of magnolol (25.0–100.0 mg/kg bw) produced a decrease in colon temperature, an increase in foot skin temperature, a decrease in metabolic rate and a decrease in the endogenous release of 5-hydroxytryptamine in the rat hypothalamus. Depletion of 5-hydroxytryptamine in rat brain, produced by intracerebroventricular pretreatment with 5,7-dihydroxytryptamine, attenuated the magnolol-induced hypothermia and cutaneous vasodilation, and decreased metabolism (*37*).

Muscle relaxation effects

Intraperitoneal administration of an ether extract of the crude drug, at a dose of 1.0 g/kg bw induced a loss of the righting reflex 30 minutes after injection. In mice, intraperitoneal administration of magnolol (50.0–500.0 mg/kg bw) produced sedation, ataxia, muscle relaxation and a loss of righting reflex. Intraperitoneal administration of magnolol and honokiol (50.0 mg/kg bw) to young chicks suppressed spinal reflexes (*38–40*).

Toxicity

The median lethal dose after intraperitoneal injection of a decoction is 6 g/kg bw in mice, and the median lethal dose is 4.25 g/kg bw in cats after intravenous administration (*19*, *34*).

Clinical pharmacology

No information was found for single-entity preparations of Cortex Magnoliae.

Contraindications
Hypersensitivity or allergy to the plant material.

Warnings
No information was found.

Precautions
General
No information was found.

Drug interactions
None reported.

Drug and laboratory test interactions
None reported.

Carcinogenesis, mutagenesis, impairment of fertility
An aqueous extract of the bark was not mutagenic in the Ames test in *Salmonella typhimurium* strains TA98 and TA100 at concentrations up to 40.0 mg per agar plate (*41*). Magnolol at concentrations of 10^{-9} and 10^{-7} M reduced ferrous sulfate-induced lipid peroxidation and reduction in sperm motility and protected sperm from lipid peroxidation in vitro (*42*).

Pregnancy: teratogenic effects
None reported.

Pregnancy: non-teratogenic effects
Due to a lack of safety data the use of the crude drug during pregnancy is not recommended.

Nursing mothers
Due to a lack of safety data the use of the crude drug during breastfeeding is not recommended.

Paediatric use
Due to a lack of safety data the use of the crude drug in children under the age of 12 years is not recommended.

Adverse reactions
No information was found.

Dosage forms

Crude drug, decoction. Store in a tightly sealed container away from heat and light (*2*).

Posology

(Unless otherwise indicated)

For oral administration: 3–9 g of crude drug for decoction daily in divided doses (*2*, *19*).

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Herba Millefolii

Definition

Herba Millefolii consists of the whole or cut, dried flowering tops (*1*, *2*) or aerial parts collected during the flowering season (*3*, *4*) of *Achillea millefolium* L. (Asteraceae).

Synonyms

Achillea borealis Bong., *A. lanulosa* Nutt., *A. magna* auct., *A. millefolium* ssp. *borealis* (Bong.) Breitung., *A. millefolium* ssp. lanulosa (Nutt.) Piper, *A. millefolium* var. *occidentale* DC (*5*).

Selected vernacular names

Achillée, achillenkraut, amelotu, artemisia bastarda, Bauchwehkraut, berbe militaris, biranjasif, bloodwort, bumadaran, carpenter's grass, carpenter's weed, chipmunk grass, centofoglie, cickafark, ciento en rama, common yarrow, daun seribu, dog daisy, egel tologch ovs, erba da carpentierir, erba da falegnam, erva d'o marchese, flor de la pluma, gandana, gordoloba, green arrow, herbe au charpentier, herbe de millefeuille, hezarbarg, Jungfraukraut, Katzenkraut, knight's milfoil, mil de tama, mil en rama, mil flores, mil hojas, milefolio, milfoil, millefolium, milenrama, nosebleed, old man's pepper, oum alf ouraka, pharange, saigum, sanguinary, sataraatyoutas, Schafgarbe, Schafgarbenkraut, seiyonokogiriso, seiyounokogirisou, sneezeweed, soldier's milfoil, stratictes, tansy, thou alf ouraka, thousand leaf, thousand leaf grass, thousand seal, thousand weed, trava tysyachelistnik, troneto, umm alf waraqah, western yarrow, wound wort, yarrow, yerba de carpintero, yerba de la muela (*2*, *6–10*).

Geographical distribution

Native to Asia, Europe and North America, now widely distributed and cultivated in the temperate regions of the world (*2*, *7*, *8*, *11*, *12*).

Description

A perennial herb, 30–90 cm in height, with aromatic odour and greyish-green colour from the numerous small hairs; stem angular. Leaves green

or greyish-green, faintly pubescent on the upper surface and more pubescent on the lower surface, 2–3 pinnately divided with linear lobes and a finely pointed whitish tip, alternate, clustered at the base of the stem. Flowering heads (capitula) in a flat-topped corymb (3–5 cm in diameter), small, pedunculate, varying in colour from white to pink, magenta and red; involucral bracts in few rows, the outer somewhat shorter than the inner, with a scarious margin. Outer florets in each capitulum usually 5, female, ligulate with more or less 3-dentate, patent ligules; inner florets hermaphrodite, 5-lobed, with compressed corolla tube and a receptacle scale at the base. Fruit a compressed achene, oblong or obovate, without pappus (*1*).

Plant material of interest: dried flowering tops and aerial part

General appearance

Flowering tops: Leaves green or greyish-green, faintly pubescent on the upper surface and more pubescent on the lower surface, 2–3 pinnately divided with linear lobes and a finely pointed whitish tip. The capitula are arranged in a corymb at the end of the stem. Each capitulum (3–5 cm in diameter) consists of the receptacle, usually 4 or 5 ligulate ray-florets and 3–20 tubular disc florets. The involucre consists of 3 rows of imbricate lanceolate, pubescent green bracts arranged with a brownish or whitish, membranous margin. The receptacle is slightly convex, and in the axillae of paleae, bears a ligulate ray floret with a 3-lobed, whitish or reddish ligule and tubular disc florets with a radial, 5-lobed, yellowish or light brownish corolla. The pubescent green, partly brown or violet stems are longitudinally furrowed, up to 3 mm thick with a light-coloured medulla (*1*).

Aerial part: Stems rounded, pubescent, furrowed, usually unbranched, 40 cm or more in length, distinctly woolly, pale green, sometimes purplish. Lanceolate leaves, up to 15 cm in length and 3 cm in width, 2 to 3 pinnate with the ultimate segments linear and subulate, pale greyish-green and covered with long white hairs; lower leaves with a short petiole, upper leaves sessile, often with two or three small axillary leaves at the base. Flowers numerous, in dense terminal corymbs, each capitulum about 3–5 cm in diameter with an ovoid involucre composed of 3 rows of imbricate lanceolate, pubescent green bracts arranged with a brownish or whitish, membranous margin; 4 or 5 white, pink or reddish ligulate ray-florets and 3–20 white or cream tubular disc florets; achenes 2 mm long, shiny, greyish-brown, slightly curved (*1*, *3*, *4*).

Organoleptic properties

Odour: slightly aromatic; taste: bitter, faintly aromatic (*3*, *4*, *7*).

Microscopic characteristics

Aerial part: Stem shows epidermal cells axially elongated with occasional anomocytic stomata and a faintly striated cuticle; abundant covering and scattered glandular trichomes; cortex narrow, parenchymatous with several layers of collenchyma in the ridges; numerous vascular bundles, arranged in a ring in transverse section, each with a small group of phloem and a wide cap of thick-walled, lignified pericyclic fibres; parenchymatous cells of outer pith lignified and pitted, those of the central region unlignified and sometimes collapsed in older stems forming a hollow. Leaf cells isobilateral, with palisades composed of 1–3 layers; upper and lower epidermal cells with sinuous anticlinal walls and numerous anomocytic stomata; abundant covering trichomes and scattered glandular trichomes occurring on both epidermises. Flower epidermal cells consisting of bracts, longitudinally elongated, thin-walled, filled with dark brown striated pigment, scattered covering trichomes and occasional stomata; the inner central region composed of elongated cells with lignified and finely pitted walls. Corolla of the ray floret with the epidermis of the ligule composed of wavy-walled cells with rounded papillae; corolla of the disc floret composed of rectangular cells with moderately thickened walls; numerous small cluster crystals of calcium oxalate occur in both ray and disc florets. Pollen grains spherical, 30–35 µm in diameter, with a spiny exine and 3 distinct pores (*4*).

Powdered plant material

Flowering tops: Green or greyish-green. Fragments of stems, leaves, and bracts bearing rare glandular trichomes with a short stalk and a head formed of 2 rows of 3–5 cells enclosed in a bladder-like membrane and uniseriate covering trichomes consisting of 4–6 small, more or less isodiametric cells at the base and a thick-walled, often somewhat tortuous terminal cell, 400–1000 µm in length; fragments of the ligulate corolla with papillary epidermal cells; small-celled parenchyma from the corolla tubes containing cluster crystals of calcium oxalate; groups of lignified and pitted cells from the bracts; spherical pollen grains, about 30 µm in diameter, with 3 germinal pores and spiny exine; groups of sclerenchymatous fibres and small vessels with spiral or annular thickening, from the stem (*1*).

Aerial part: Greyish-green powder with epidermal fragments of stem and leaf with abundant covering trichomes and less numerous glandular trichomes, the covering trichomes frequently broken off and occurring scattered; groups of thick-walled, lignified fibres from the pericycle and xylem, those of the xylem sometimes associated with small vessels with spiral or annular thickening; lignified, pitted parenchyma from the pith;

dark brown fragments of the membranous margins of the bracts and groups of lignified and pitted elongated cells from the central region; occasional fragments of the papillose epidermis of the ligulate florets; small-celled parenchyma containing cluster crystals of calcium oxalate; pollen grains with a spiny exine (*4*).

General identity tests

Macroscopic and microscopic examinations (*1*, *3*, *4*), and thin-layer chromatography (*1*).

Purity tests

Microbiological

Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*13*).

Foreign organic matter

Flowering tops: not more than 5% of stems with a diameter greater than 3 mm and not more than 2% of other foreign matter (*1*).

Aerial part: not more than 2% (*4*).

Total ash

Flowering tops: not more than 10.0% (*1*).
Aerial part: not more than 10% (*4*).

Acid-insoluble ash

Flowering tops: not more than 2.5% (*1*).
Aerial part: not more than 2.5% (*4*).

Water-soluble extractive

Aerial part: not less than 15.0% (*4*).

Loss on drying

Flowering tops: not more than 12.0% (*1*).
Aerial part: not more than 13% (*3*).

Pesticide residues

The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*1*). For other pesticides, see the *European pharmacopoeia* (*1*) and the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*13*) and pesticide residues (*14*).

Heavy metals

For maximum limits and analysis of heavy metals, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*13*).

Radioactive residues

Where applicable, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*13*).

Chemical assays

Flowering tops: not less than 0.2% (v/w) of essential oil calculated on the basis of dried weight; and not less than 0.02% of proazulenes expressed as chamazulene by a combination of steam distillation and spectroscopic analysis (*1*).

Aerial part: not less than 0.1% (v/w) of essential oil determined by steam distillation (*3*).

Major chemical constituents

Contains 0.2–1.0% of essential oil. Being a chemically polymorphic aggregate plant species, the chemical constitution depends on the number of chromosomes present. Diploid and tetraploid plants contain proazulene sesquiterpenes, which when exposed to heat will be transformed to coloured azulenes, including chamazulene (up to 25%) and achillicin. Other major constituents in tetraploid plants include β-pinene (23%), α-pinene (5%) and caryophyllene (10–22%). Hexaploid plants are azulene sesquiterpene-free, and contain approximately 50% mono- and sesquiterpenes, many of which are in the oxidized form, as well as camphor (18%), sabinene (12%), 1,8-cineol (10%) and β-pinene (9%), among other constituents. Octaploid plants contain approximately 80% oxygen-containing monoterpenes, with linalool being the major constituent. Among the non-essential-oil constituents are flavonoids, coumarins and tannins (*6*, *7*, *9*, *11*, *15*). The structures of representative mono- and sesquiterpenes are presented below.

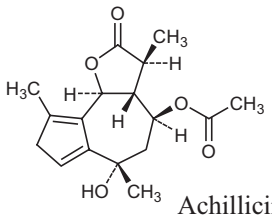
Medicinal uses

Uses supported by clinical data

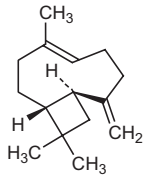
None.

Uses described in pharmacopoeias and well established documents

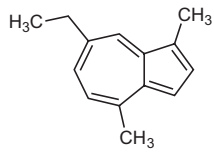
Orally for loss of appetite, common cold, dyspeptic ailments such as mild spastic discomfort of the gastrointestinal tract, as a choleretic and for the



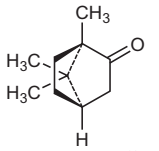
Achillicin



Caryophyllene



Chamazulene



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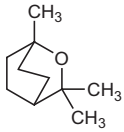
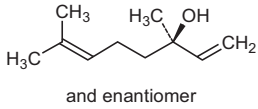
Camphor



1,8-Cineole



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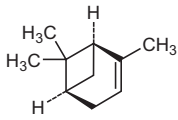
Linalool



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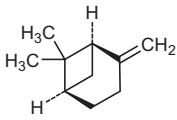
α-Pinene



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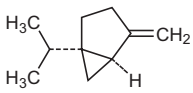
β-Pinene



and enantiomer

Sabinene

treatment of fevers (*6*, *12*, *16*). Externally for skin inflammation and wounds (*6*).

Externally as a sitz bath for treatment of painful, cramp-like conditions due to menstrual disorders (*12*).

Uses described in traditional medicine

Orally as an emmenagogue, eyewash, haemostat, laxative, sleep aid, stimulant tonic, and to treat baldness, prostatitis and vertigo (*8*, *9*, *15*, *17*, *18*).

Used externally for the treatment of haemorrhoids, haematoma and burn injuries (*19*).

Pharmacology

Experimental pharmacology

Note: While the flowering tops of the plant are official in the *European pharmacopoeia* 2005 (*1*), much of the research on the pharmacology of this plant has been performed using the aerial parts of the plant, which include the flowering tops. These data have been included and designated as coming from studies conducted on the aerial parts, but their direct applicability to the flowering tops needs to be further investigated.

Antibacterial activity
A 50% ethanol extract of the flowers inhibited the growth of *Shigella dysenteriae*, but not that of *Escherichia coli* or *Salmonella enteritidis*, in vitro at a concentration of 50 µl/agar plate (*20*). A methanol extract of the aerial parts inhibited the growth of 18 clinical strains of *Helicobacter pylori* in vitro, with a minimum inhibitory concentration of 50 µg/ml (*21*).

Anticonvulsant activity
Intraperitoneal injection of a 95% ethanol extract of the aerial parts to mice, at a dose of 2.0–4.0 ml/kg body weight (bw), had anticonvulsant activity against supramaximal electroshock- and corazol-induced convulsions, but was not effective against strychnine-induced convulsions (*22*).

Anti-inflammatory activity
In a study in mice, intraperitoneal injection of a fraction from an aqueous extract of the flower heads, at a dose of 40.0 mg/kg bw, inhibited yeast-induced pedal oedema (*23*). Intragastric administration of an 80% ethanol extract of the aerial parts to rats, at a dose of 100.0 mg/kg bw, inhibited carrageenan-induced pedal oedema by 29% (*24*). External application of a methanol extract of the aerial parts to mice, at a dose of 1.0 mg/ear, had weak anti-inflammatory effects (*25*). An aqueous extract of the aerial parts did not inhibit prostaglandin synthesis in microsomes at a concentration of 0.2 mg/ml (*26*). Santamarin, a sesquiterpene lactone from the crude drug, moderately inhibited the transcription of nuclear factor-kappa-beta, a protein that regulates the transcription of inflammatory mediators such as the cytokines and chemokines, at a concentration of 100 µM (*27*).

Antioxidant activity
Chamazulene, an artefact constituent of the aerial parts, inhibited cell membrane lipid peroxidation induced by Fe2+/ascorbate as assessed in the 2-thiobarbituric acid reactive assay. Chamazulene inhibited lipid peroxidation in a concentration- and time-dependent manner, with a median inhibitory concentration of 18 µM. It also inhibited the autoxidation of dimethylsulfoxide (33 mM) by 76% at 25 mM, and had a weak capacity to interact with 2,2-diphenyl-1-picrylhydrazyl (*28*).

Antipyretic activity
Oral administration of a hot aqueous extract or the juice of the aerial parts of the plant to rabbits, at a dose of 25 and 55 g/kg bw, respectively, reduced body temperature, while the 95% ethanol extract was not active (*29*).

Antispasmodic activity

An aqueous or methanol extract of the aerial parts of the plant (concentration not stated) inhibited contractions of rabbit small intestines in vitro (*30*).

Antiviral activity

A 50% methanol extract of the aerial parts inhibited HIV-1 reverse transcriptase in vitro at a concentration of 10% of the nutrient medium (*31*). Intraperitoneal administration of a hot-water extract of the dried flowers and leaves of the plant to mice (dose not stated) was active against tick-borne viral encephalitis (*32*).

Toxicology

Intraperitoneal administration of an aqueous extract of the aerial parts to rats had a median lethal dose of 1.5 g/kg bw (*33*). Intragastric or subcutaneous administration of an aqueous extract of the flowers to mice had a median lethal dose of > 1 g/kg bw (*33*).

Clinical pharmacology

Oral administration of a 70% ethanol extract of the flowers (dose not stated) increased the secretion of gastric juice in healthy volunteers by 178% (*16*). No further information on this study was available.

Adverse reactions

Numerous reports of allergic contact dermatitis have been published (*33–39*). In clinical testing, product formulations containing 2% of extracts of the crude drug were generally not irritating. In provocative testing, patients reacted to a Compositae mix that contained the crude drug, as well as to the crude drug alone. In clinical testing, a formulation containing 0.1% yarrow extract (propylene glycol and water) was not a sensitizer in a maximization test and alcoholic extracts of aerial parts of *A. millefolium* did not produce a phototoxic response (*33*).

A 5-year follow-up (1985–1990) of patients who were sensitive to Compositae showed that more than 50% reacted when tested with an ether extract of the plant, indicating cross-sensitivity (*35*). However, exacerbation of the patch test sites by irradiation with UV light was not observed in any of the tested patients. One guaianolide compound, with a peroxide-bridged cyclopentane ring and an α-methylene-γ-butyrolactone structure, named α-peroxyachifolide, has been isolated from the flowers and appears to be responsible for the allergic contact dermatitis (*35*, *37*).

Therefore, direct contact with the crude drug or its preparations may cause hypersensitivity reactions of the skin or mucosa, such as rash, formation of vesicles and pruritus, in sensitive individuals.

Contraindications

Hypersensitivity to the plant and other Asteraceae (Compositae) (*12*, *40*, *41*). Gastric and duodenal ulcer, occlusion of the bile duct and gallbladder disease (*12*).

Due to the traditional use of the drug as an emmenagogue, it is contraindicated during pregnancy (*9*).

Warnings

Patients presenting with hypersensitivity or allergic reactions that include the formation of vesicles should stop treatment with Herba Millefolii immediately (*40*). If signs of hypersensitivity reaction reappear upon further use, the crude drug should not be used again.

Precautions

Carcinogenesis, mutagenesis, impairment of fertility

A tincture of the crude drug was not mutagenic in the Ames test at a concentration of 160 µl/disc in *Salmonella typhimurium* strains TA98 and TA100. Metabolic activation had no effect on the results (*33*). An infusion of the aerial parts was tested for genotoxicity in the wing somatic mutation and recombination test (SMART) which makes use of the two recessive wing cell markers, multiple wing hairs (mwh) and flare (flr) on the left arm of chromosome 3 of *Drosophila melanogaster*. Three-day-old larvae, transheterozygous for these two markers, were fed the beverage (an infusion of *Achillea millefolium* (20 g/100 ml water) cooled and used immediately for the larval experiments) at different concentrations and for different feeding periods using *Drosophila* instant medium. Somatic mutations or mitotic recombinations induced in the cells of the wing imaginal discs gave rise to mutant single or twin spots on the wing blade of the emerging adult flies showing either the mwh phenotype and/or the flr phenotype. An infusion of *Achillea millefolium* was weakly genotoxic (*42*).

The results of previous investigations assessing the effect of the crude drug on reproduction have been contradictory. In one study, the addition of the plant to the feed of rats, at a concentration of 25–50% w/w, suppressed the induction of estrus (*43*). However, oral administration of an extract of the leaves to rats did not alter the time of first mating, fertility or litter size (*44*). The effect of a 96% ethanol extract (200 mg/kg bw per day, intraperitoneally, for 20 days) and an 80% ethanol extract (300 mg/kg bw per day, orally, for 30 days) of the flowers on the spermatogenesis of Swiss mice was assessed by examining morphological characteristics with light and electron microscopes. Neither dose caused a significant dif-

ference in body weight gain or in the weight of the testes and seminal vesicles. The alterations observed were exfoliation of immature germ cells, germ cell necrosis, and seminiferous tubule vacuolization. Animals treated with the extracts had an increased number of metaphases in the germ epithelium that might be due to cytotoxic substances or substances stimulating cell proliferation (*45*).

Pregnancy: Non-teratogenic effects
See Contraindications.

Nursing mothers
Due to the lack of safety data the crude drug should not be used by breastfeeding mothers.

Paediatric use
Due to the lack of safety data the crude drug should not be used in children under the age of 12 years.

Other precautions
No information was found.

Dosage forms
Crude drug, extracts, fluidextract, infusions, succus (pressed juice from fresh herb) and tinctures.

Posology
(Unless otherwise indicated) (*12*)
Internal: 4.5 g of cut herb (flowering top) per day, or 3.0 g cut flowers for teas (infusions) and other Galenical preparations; pressed juice of freshly harvested herb.
Infusion: 1–2 g in 150 ml boiled water for 10–15 minutes, three times daily between meals.
Succus (pressed juice from fresh herb): 5 ml (1 teaspoon), three times daily between meals.
Fluidextract 1:1 (g/ml): 1–2 ml, three times daily between meals.
Tincture (1:5 g/ml): 5 ml, three times daily between meals.
External: sitz bath: 100 g per 20 litres of warm or hot water (*12*).

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Fructus Momordicae

Definition

Fructus Momordicae consists of the fresh (*1*) or the dried fruits of *Momordica charantia* L. (Cucurbitaceae).

Synonyms

Momordica balsamina Blanco, *M. chinensis* Spreng., *M. elegans* Salisb., *M. indica* L. (*2*, *3*).

Selected vernacular names

African cucumber, amargoso, ampalaya, art pumpkin, asorosi, assorossi, balsam apple, balsam pear, Balsambirne, balsamina, balsamino, ban kareli, baramasiya, barbof, bitter cucumber, bitter gourd, bitter melon, bitter pear melon, bobobo, broomweed, calaica, caprika, carailla, carilla, cerasee, concombre, condiamor, coraillie, cun de amor, cundeamor, ejirin, embusabusu, eyezom, futoreishi, goya, haagalakaayi, hagalakai, 'ha, 'hail, kaakara kaaya, kabiral, kaippa, kakara, kakayi, kakiral, kakle, kakral, karala, kãravella, kãravellí, karawila, karela, karla, karlara, karolla, kathila, khoor qua, khyar carilla, kokouli, kugua tea, kuguazi, lenzaa, lumba-lumba, lumbuzi, ma ra, machete, maha, maiden apple, maiden's blush, manamat, mange kuli, mara, mara khee nok, margoze, mbosa, mbunbulu, melao de sao caetano, meleni, mexicaine, miniklalasni, momotica, mreah, muw, nagareishi, nania nania, nara cheen, nguene, nigauri, nyanyra, nyinya, okookoo, pagel, paharkai, palia, panaminik, papari, papayilla, papilla, paria, pavakka, pavel, 'phak, paroka, pavackkai, pavakkachedi, pepino montero, peria, periya laut, phakha, pom kouli, pomme nerveille, pomme z'indiens, pomme-cooli, qara à mor, qisaul-barri, quisaul-barri, serimentok, seripupa, saga, sail, salara, sorosi, sorrow see, sushavi, tasplira, tofah al, uchhe, ulhimar, varivallí, wild balsam pear, wild sopro, yesquin, zague zrou (*1*, *3*, *4*, *5–9*).

Geographical distribution

Pantropical distribution and cultivated widely (*3*).

Description

A monoecious, annual climbing vine up to 5 m long. Stem 5-ridged, with simple tendrils. Leaves alternate, petiolate, broad blade ovate to suborbicular or ovate-reniform in outline, 2.5–10.0 × 3.0–12.5 cm with 3–9 deep palmate lobes quite variable in size. Unisexual solitary flowers, yellow to orange, 3.5 cm in diameter; male flowers on a 0.5–3.0 cm long peduncle bearing an apical bract up to 2.2 cm long, and 2.0–5.5 cm long pedicel; female flowers on a 0.2–5.0 cm long peduncle bearing an apical bract of up to 1.2 cm long, and pedicel 1.0–10 cm in length. Fruits, fleshy, oblong-fusiform-cylindrical, irregularly warty, usually 3–11 cm, but can be up to 45 cm in length × 2–8 cm in diameter, orange, dehiscing. Seeds, 8–16 × 4–10 × 2.5–3.5 mm, brown, testa ornamented (*3*, *7*, *10*).

Plant material of interest: fresh or dried fruits

General appearance

Fleshy, oblong-fusiform-cylindrical, irregularly warty, usually 3–11 cm, but cultivars can be up to 45 cm in length × 2–8 cm in diameter, green (young) to orange (old), dehiscing. Seeds, 8–16 × 4–10 × 2.5 × 3.5 mm, brown, testa ornamented (*3*, *7*, *10*).

Organoleptic properties

Odour: characteristic; taste: bitter (*10*).

Microscopic characteristics

To be established in accordance with national requirements.

Powdered plant material

To be established in accordance with national requirements.

General identity tests

Macroscopic examination (*3*, *7*, *10*).

Purity tests

Microbiological

Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*11*).

Foreign organic matter

None (*1*).

Total ash
Not more than 8.5% (*1*).

Acid-insoluble ash
Not more than 0.6% (*1*).

Water-soluble extractive
Not less than 28% (*1*).

Alcohol-soluble extractive
Not less than 6% (*1*).

Loss on drying
To be established in accordance with national requirements.

Pesticide residues
The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*12*). For other pesticides, see the *European pharmacopoeia* (*12*) and the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*11*) and pesticide residues (*13*).

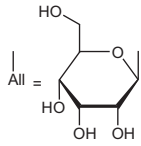
Heavy metals
For maximum limits and analysis of heavy metals, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*11*).

Radioactive residues
Where applicable, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*11*).

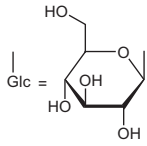
Chemical assays
To be established in accordance with national requirements.

Major chemical constituents
A variety of primary and secondary constituents have been reported in the fruits of *M. charantia*. Characteristic constituents include sterols, triterpenes and bioactive proteins (α- and β- momorcharins; sterols (e.g. daucosterol); triterpenes, goyaglycosides a–h, goyasaponins I–III, cucurbitacins and their glycosides such as momordicosides E1, F1, F2, F–K. Gallic acid, gentisic acid, catechin, chlorogenic acid and epicatechin are the most frequently found low-molecular-weight phenolics (*5*, *14–20*, *22*, *23*).

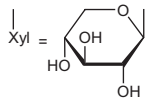
Structures of representative major constituents are presented below.



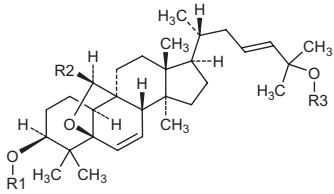
β-D-allopyranosyl



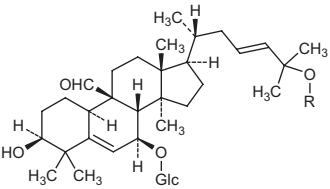
β-D-glucopyranosyl



β-D-xylopyranosyl

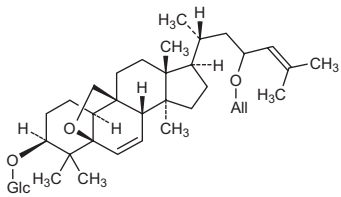


	R1	R2	R3
Momordicoside I	Glc	H	H
Momordicoside F2	All	H	H
Momordicoside F1	Glc	H	CH_3
Momordicoside G	All	H	CH_3
Goyaglycoside a	Glc	OCH_3	H
Goyaglycoside b	All	OCH_3	H
Goyaglycoside c	Glc	OCH_3	CH_3
Goyaglycoside d	All	OCH_3	CH_3
Goyaglycoside e	All	H	Glc
Goyaglycoside g	All	OCH_3	Glc

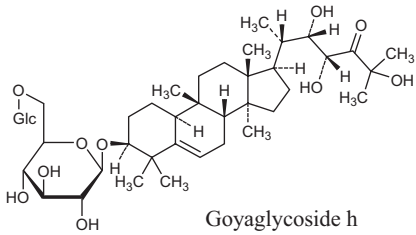


Momordicoside K R = CH_3

Momordicoside L R = H



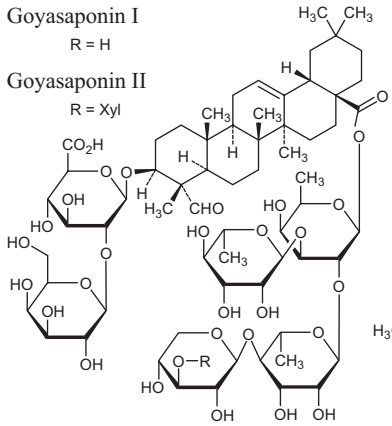
Goyaglycoside f



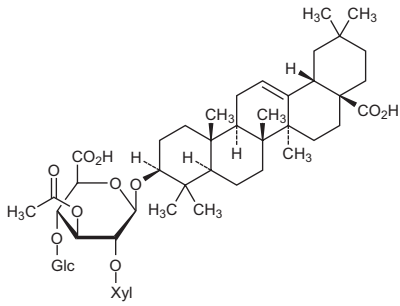
Goyaglycoside h

Goyasaponin I
R = H

Goyasaponin II
R = Xyl



Goyasaponin III



Medicinal uses

Uses supported by clinical data

None.

Uses described in pharmacopoeias and well established documents

Used as an antidiabetic agent, an emetic, a laxative and a tonic (*1*, *6*).

Although a number of case-reports and pilot studies have suggested that various preparations of the fruit may improve glucose tolerance, fasting blood glucose levels and glycosuria (*24–31*), none of the trials were randomized or blinded. Furthermore, the dosage, toxicity and adverse events have not been systematically assessed. Adequately powerful, randomized, placebo-controlled trials are needed to properly assess the safety and efficacy of preparations of the fruit before therapeutic recommendations can be made.

Uses described in traditional medicine

Used to treat anaemia, arthritis, colds, fever, gout, infertility, kidney stones, peptic ulcers, stomach ache and worms (*5*, *7*).

Pharmacology

Experimental pharmacology

Effects on streptozotocin-induced diabetes

The effects of a dried ethanol extract of the fruit pulp (1 kg fruit pulp minus seeds in 1500 ml 95% ethanol) were investigated in a normal glucose-primed rat model and streptozotocin-induced diabetic rats. Oral administration of the extract, at a dose of 500.0 mg/kg body weight (bw), reduced the plasma glucose levels in normal rats by 10–15% after 1 hour. In rats with streptozotocin-induced diabetes, the extract significantly improved the oral glucose tolerance ($p < 0.002$) and reduced plasma glucose by 26% at 3.5 hours while metformin caused a 40–50% reduction after 1, 2 and 3.5 hours (*32*).

A diet containing the crude drug (30.0 g/kg bw) was fed to mice with streptozotocin-induced diabetes daily for 21 days. The serum glucose concentration of mice fasting for 12 hours and 2 hours after a meal was determined on the 21st day of the experiment. The results demonstrated that the serum glucose levels of diabetic mice were significantly higher than those of the mice in the normal control group ($p < 0.01$). After treatment for 21 days, the serum glucose concentrations of the treated diabetic mice were significantly lower than those of the mice in the streptozotocin-induced diabetic control group ($p < 0.01$), but were still higher than the normal control group (*33*).

Alcohol- and aqueous extracts of the fruit pulp (1:1) (50.0, 100.0 and 200.0 mg/kg bw per day) were evaluated in a pilot study (plasma glucose > 180 mg/dl, 21 days), a chronic study in alloxanized rats (plasma glucose > 280.0 mg/dl, 120 days) and streptozotocin-treated mice (plasma glucose > 400.0 mg/dl, 60 days). The maximum antihyperglycaemic effect occurred with an aqueous extract at week 3, at a dose of 200.0 mg/kg bw per day. In chronic alloxanized rats, treatment with the aqueous extract led to a reduction of 64.33%, 66.96%, 69.7% and 70.53% in plasma glucose levels after 1, 2, 3 and 4 months of treatment, respectively. In mice with chronic streptozotocin-induced diabetes, treatment with the aqueous extract led to a mean reduction of 15.37%, 18.68% and 22.86% in plasma glucose levels on days 40, 50 and 60, respectively (*34*).

Another similar study that examined the hypoglycaemic potency of the crude drug in rats with streptozotocin-induced diabetes found no effect (*35*). In this investigation the animals were maintained on a semi-synthetic diet containing 0.5% freeze-dried bitter gourd powder for 6 weeks. The excretion of glucose, protein, urea and creatinine was monitored throughout the experiment. Plasma glucose, albumin, urea and cholesterol were analysed at the end of the experiment. No beneficial effects were observed (*35*).

The anti-hyperglycaemic effects of three extracts of the crude drug were assessed in rats. Three different aqueous extracts of powdered fresh or dried whole fruits were prepared and orally administered to rats with streptozotocin-induced diabetes. An aqueous extract of the powdered fresh unripe whole fruits was the most active, and, at a dose of 20.0 mg/kg bw, reduced fasting blood glucose by 48%, an effect comparable to that of glibenclamide. The aqueous extract was tested for nephrotoxicity, hepatotoxicity and elevation of liver enzymes and showed no signs of nephrotoxicity or hepatotoxicity (*36*).

The antidiabetic activity of an aqueous extract of the fruit was assessed in KK-Ay mice with type 2 diabetes with hyperinsulinaemia. The extract reduced the blood glucose of KK-Ay mice 3 weeks after oral administration ($p < 0.01$) and also significantly lowered the serum insulin of KK-Ay mice under similar conditions ($p < 0.01$). However, the extract did not affect the blood glucose in normal mice. In KK-Ay mice treated with the extract, blood glucose was significantly decreased in an insulin tolerance test. Moreover, the muscle content of facilitative glucose transporter isoform 4 (GLUT4) protein in the plasma membrane fraction of muscle significantly increased in the mice that received the extract orally when compared with that of the controls ($p < 0.01$) (*37*). In a follow-up study, the antidiabetic activity of the extract combined with exercise was investi-

gated in KK-Ay mice. The extract in combination with exercise reduced the blood glucose concentration of KK-Ay mice 5 weeks after oral administration daily for 5 weeks ($p < 0.001$), and also significantly lowered the plasma insulin of KK-Ay mice under similar conditions ($p < 0.01$). The blood glucose concentration following treatment with the extract plus exercise was lower than that following treatment with the extract only or exercise only, 5 weeks after the administration. Administration of the extract combined with exercise decreased blood glucose in a glucose tolerance test (*38*).

Effect on insulin resistance

In a study in rats, the effect of the crude drug on insulin resistance, a major contributor to the development of hyperglycaemia, was assessed in vivo. The effects of different doses (100.0, 200.0 and 400.0 mg per day) of alcohol extracts or aqueous extracts of the fruits (1 kg of fruit to 500 ml ethanol) on the metabolic parameters (body weight and serum glucose insulin and triglyceride levels) of rats fed fructose were studied. Feeding fructose for 15 days increased serum glucose and insulin levels markedly and triglyceride levels marginally when compared with the controls (75.46 versus 55.59 mg/dl, 6.26 versus 15.04 mg/dl and 50.93 versus 41.1 mg/dl, respectively). Treatment with 400.0 mg/day of aqueous extract (1 kg in 500 ml water) for 15 days substantially reduced hyperglycaemia and hyperinsulinaemia induced by a diet high in fructose (*39*).

The antidiabetic activity of an aqueous extract of the fruit was investigated in KK-Ay mice, an animal model having type 2 diabetes with hyperinsulinaemia. Intragastric administration of an aqueous extract of the fruit reduced the blood glucose of KK-Ay mice after 3 weeks of intragastric administration ($p < 0.01$) and also significantly lowered the serum insulin ($p < 0.01$). However, the extract did not affect the blood glucose in normal mice. Blood glucose in treated KK-Ay mice was significantly decreased in an insulin tolerance test ($p < 0.01$) (*37*).

The activity of the fruit juice was assessed in vitro on streptozotocin-treated RIN cells and isolated islet cells, and in vivo in mice. It was found that feeding juice caused a reduction in streptozotocin-induced hyperglycaemia in mice. The juice reduced the streptozotocin-induced lipid peroxidation in pancreas of mice, RIN cells and islets. It also reduced the streptozotocin-induced apoptosis in RIN cells indicating the mode of protection of the juice on RIN cells, islet cells and pancreatic beta-cells (*40*). The activity of the fruit juice on the distribution and number of alpha, beta and delta cells in the pancreas of rats with streptozotocin-induced diabetes was determined using immunohistochemical methods. The results suggested that there was a significant ($p < 0.004$) increase in the

number of beta cells in treated animals, as compared with untreated diabetic rats, but their number was still significantly smaller than that obtained for normal rats. There was also a significant ($p < 0.006$) increase in the number of delta cells in rats with streptozotocin-induced diabetes compared to non-diabetic rats, but the number of alpha cells did not change. These data indicate that oral administration of the crude drug may renew the beta cells in rats with streptozotocin-induced diabetes or alternatively may permit the recovery of partially destroyed beta cells (*41*).

The effect of the fruit juice on rats fed a hyperinsulinaemic high-fat diet was investigated. In a dose–response study (0.375, 0.75 and 1.5% freeze-dried juice administered in rations), oral glucose tolerance was improved in rats fed a high fat (30%) diet supplemented with freeze-dried juice at a dose of 0.75% or higher ($p < 0.05$). At the highest dose, treated rats had lower energy efficiency ($p < 0.05$) and tended to have a lower visceral fat mass ($p = 0.10$). In a subsequent experiment, rats habitually fed a high fat diet either continued to consume the diet or were switched to a high fat + fruit juice, low fat (7%), or low fat + fruit juice (0.75%) diet for 7 weeks. Rats which had been switched to the high fat + fruit juice diet gained less weight and had less visceral fat than those fed the high fat diet ($p < 0.05$). Supplementation of the high fat diet with the crude drug improved insulin resistance, lowered serum insulin and leptin, but raised free fatty acid concentration in the serum ($p < 0.05$) (*42*).

Antihypercholesterolaemic activity

A long-term (10-week) feeding experiment measured the effect of a fruit extract (10 ml 100% fruit juice/kg bw) on blood plasma and tissue lipid profiles in normal rats and rats with streptozotocin-induced type 1 diabetes. The results showed a significant ($p < 0.05$) increase in plasma non-esterified cholesterol, triglycerides and phospholipids in rats with streptozotocin-induced diabetes, accompanied by a decrease in high-density lipoprotein-cholesterol. A moderate increase in the plasma lipid peroxidase product, malondialdehyde, and about a twofold increase in kidney plasma lipid peroxidase was also observed. The treatment of diabetic rats with the fruit extract over a 10-week period returned these levels to normal. In addition, the extract exhibited an inhibitory effect on membrane plasma lipid peroxidase under in vitro conditions (*43*).

The effects of a freeze-dried powder of the fruit on serum glucose levels and lipid parameters of the serum and liver were studied in rats fed diets supplemented with cholesterol and without cholesterol. For 14 days, rats were fed the diets either containing the freeze-dried powder at 0.5, 1 or 3% without added dietary cholesterol (experiment 1) or containing 1% powder with or without 0.5% cholesterol and 0.15% bile acid (ex-

periment 2). Dietary intake of powdered crude drug resulted in a consistent decrease in serum glucose levels in rats fed cholesterol-free diets, but not in those fed cholesterol-enriched diets, although no dose–response was noted. The crude drug had no effect on serum lipid parameters, except for high-density lipoprotein-cholesterol levels, which were elevated, indicating an anti-atherogenic activity. In addition, the crude drug caused a marked reduction of hepatic total cholesterol and triglyceride levels both in the presence and absence of dietary cholesterol (*44*).

Antiviral activity

MAP30 (Momordica anti-HIV protein of 30 kDa), isolated from the plant material, is capable of inhibiting infection of HIV type 1 (HIV-1) in T lymphocytes and monocytes as well as replication of the virus in infected cells. An investigation of the effect of MAP30 on HIV-1 integrase suggests that the protein exhibits a dose-dependent inhibition of HIV-1 integrase. In the presence of 20 ng of viral substrate, 50 ng of target substrate, and 4 µM integrase, total inhibition was achieved at equimolar concentrations of the integrase and the antiviral proteins, with a median effective concentration of 1 µM (*45*).

A protein (MRK29), isolated from the fruit and seed, inhibited HIV-1 reverse transcriptase by 50% at a concentration of 18 µg/ml. MRK29, at a concentration of 0.175 µg/ml, reduced viral core protein, p24, expression in HIV-infected cells by 82%. MRK29 also produced a 3-fold increase in tumour necrosis factor activity (*46*).

Xenobiotic metabolism

The effects of a fruit juice (10 ml of 100% pure juice) on streptozotocin-induced diabetes on tissue-specific cytochrome P450 (CYP) and glutathione-dependent xenobiotic metabolism was investigated in rats. Results demonstrated that rats with streptozotocin-induced diabetes exhibited an increase (35–50%) in cytochrome P4504A-dependent lauric acid hydroxylation in liver, kidney and brain. About a twofold increase in cytochrome P4502E-dependent hepatic aniline hydroxylation and a 90–100% increase in cytochrome P4501A-dependent ethoxycoumarin-*O*-deethylase activities in kidney and brain was also observed. A significant increase (80%, $p < 0.01$) in aminopyrene *N*-demethylase activity was observed only in the kidney of rats with streptozotocin-induced diabetes, and a decrease was observed in the liver and brain of rats with streptozotocin-induced diabetes. A significant increase (77%) in NADPH-dependent lipid peroxidation (plasma lipid peroxidase) in kidney of diabetic rats was also observed. A marked decrease (65%) in hepatic glutathione content and glutathione S-transferase activity and an increase (about twofold) in brain glutathione

and glutathione S-transferase activity was observed in diabetic rats treated with the extract. Chronic feeding of rats with the extract reversed the effect of chronic diabetes on the modulation of both P450-dependent monooxygenase activities and glutathione-dependent oxidative stress-related lipid peroxidase and glutathione S-transferase activities (*47*).

The effect of oral feeding of the fruit juice (10 ml of 100% juice) on the hepatic cytochrome P450 and glutathione S-transferase drug-metabolizing enzymes was investigated in rats with streptozotocin-induced diabetes. Hepatic cytochrome P450 contents, ethoxycoumarin-*O*-deethylase, ethoxyresorufin-*O*-deethylase, aniline hydroxylase and aminopyrene *N*-demethylase activities were measured in control, diabetic and juice-fed animals. Diabetic rats exhibited a 50–100% increase in aniline hydroxylase and ethoxyresorufin-*O*-deethylase activities, which were reversed after being fed the juice. In addition, a decrease (17–20%) in the activities of *N*-demethylase and ethoxycoumarin-*O*-deethylase was observed in the liver of diabetic rats. Feeding the juice to the diabetic animals brought the level of *N*-demethylase close to that of control animals, while ethoxycoumarin-*O*-deethylase was further reduced to 60% of the levels in the control animals (*48*).

Toxicology

The effect of the fruit on certain key hepatic enzymes was investigated in rats. A fruit juice product was administered by gavage at a daily dose of 10 ml of pure juice/kg bw for 30 days under light ether anaesthesia while the control group received equivalent amounts of distilled water under identical conditions ($n = 10$ in each group). Serum γ-glutamyl transferase and alkaline phosphatase concentrations were found to be significantly elevated ($p < 0.001$ and $p < 0.01$–0.001, respectively), following oral administration of the fruit juice. No consistent significant histopathological changes in the liver were observed in either treatment group, although the prevalence of dilatation and/or congestion of the central vein sinusoidal system appeared to be twice as high following fruit juice treatment as in the other two groups (*49*).

Feeding of the fruit to normal adult rats at 0.02, 0.1 and 0.5% (dry weight) in a semi-synthetic diet for a period of 8 weeks had no adverse influence on the food intake, growth or organ weights of normal adult rats. The haematological parameters of the experimental rats were also normal. Serum cholesterol levels of the rats receiving 0.5% of the crude drug were significantly lower than those of the control rats (*50*).

Clinical pharmacology

Numerous case-reports and clinical studies have found that the juice, fruit, and dried fruit powder have a moderate hypoglycaemic effect. These

studies were small and were not randomized or double-blinded. Most of them assessed the anti-diabetic effects of the crude drug in both non-insulin-dependent diabetes mellitus (type II) and insulin-dependent (type I) diabetes.

In a pilot study, the efficacy of the powdered fruit was assessed in 41 patients with diabetes. The patients were treated with varying doses of the fruit for 6 months and the blood glucose level was reduced by up to 25% from a baseline value of 200 mg/dl and glycosylated haemoglobin was lowered by an average of 0.5% (*27*).

The effects of the fruit on blood glucose levels were assessed in patients with diabetes (*26*). Nineteen subjects were enrolled in the study, including 14 patients with type I or type II diabetes mellitus, and five healthy volunteers. The preparation used in this investigation was a protein isolated from the crude drug and termed "vegetable insulin". The protein extract was suspended in sterile water and administered subcutaneously at a concentration of 1.8 mg of protein per 40-unit dose. Nine of the diabetic patients received 10 units of the formulation if their fasting blood sugar was less than 180 mg/dl; 20 units when it was 180–250 mg/dl; and 30 units when it was > 250 mg/dl. Five patients with diabetes and five healthy volunteers received a placebo (sterile water). The primary endpoint was a decrease in fasting blood glucose, which was measured over 12 hours after administration. The results showed a mean decrease in serum glucose levels for the diabetic patients receiving treatment as early as 30 minutes after administration, a 21.5% decrease from mean baseline of 295 mg/dl. After 4 hours, a maximum decrease of 49.2% in serum glucose was observed, and at 12 hours a 28% drop persisted. In contrast, patients administered the placebo had a 5% decrease in serum glucose over the study period. The patients with diabetes who received treatment had substantially reduced mean baseline serum glucose concentrations, 295 mg/dl versus a concentration in the placebo group of 210 mg/dl. No blinding, randomization or statistical analyses were performed (*26*).

One investigation reports a series of case-studies in Asian patients with type II diabetes ($n = 9$), of whom eight were taking concomitant sulfonylurea drugs (*29*). After undergoing a baseline glucose tolerance test, the test was repeated after oral administration of the fruit juice (50 ml), and then again after oral ingestion, approximately 0.23 kg/day, of the fried fruit for 8–11 weeks. The glucose tolerance test performed after ingestion of the fruit showed a reduction in glucose levels of approximately 6% after 1 hour, but this finding was not statistically significant. The glucose tolerance test, done after consumption of the juice, showed a significant decrease in glucose levels of 12% after 1 hour ($p < 0.05$) and in conjunc-

tion with a glucose load resulted in a significant improvement in glucose tolerance ($p < 0.05$) without increasing insulin levels in the blood. In addition, daily consumption of the fried fruit for 8–11 weeks reduced levels of glycosylated haemoglobin by 8% from baseline (*29*). However, no randomization, blinding or proper controls were used in this study.

In another small case-report study ($n = 8$) an improvement in blood glucose tolerance and fasting blood glucose levels was observed in male and female patients (38–50 years of age) with uncomplicated type II diabetes. Patients were treated with 50 mg/kg bw of the dried fruit powder twice daily for 1 week (*25*). In addition, excretion of glucose in the urine was reduced by day 3 and was completely absent after 7 days of treatment. Mean post-treatment blood glucose levels were significantly lower than pre-treatment values. A decrease from 248 mg/dl to 155 mg/dl ($p < 0.001$) was observed in treated patients and this difference was considerably higher after the administration of 60 g of glucose. No adverse effects were reported.

In a similar case-report study, involving 12 patients with newly diagnosed type II diabetes, the effect of the crude drug on blood glucose levels was assessed. Each patient received one of two crude drug preparations. The first was an aqueous extract prepared by boiling 100 g of the crude drug in 200 ml of water until the volume was reduced to 100 ml. The second preparation was dried fruit powder, administered at a dose of 5 g three times daily. After 3 weeks of treatment the group using the powder preparation showed a reduced post-prandial blood glucose level of 25%, but the result was not statistically significant. However, in the group receiving the aqueous extract, a significant reduction (54%, $p < 0.01$) in blood glucose was observed, as well as a reduction in glycosylated haemoglobin from 8.37% to 6.95% ($p < 0.01$) (*30*).

A series of case-reports involving 18 patients with newly diagnosed type II diabetes assessed the effects of the fruit juice on blood sugar levels (*53*). The results of oral administration of 100.0 ml of the fruit juice 30 minutes prior to glucose loading for a glucose tolerance test were compared with the results of a glucose tolerance test done the previous day using water as the control test substance. Seventy-three per cent of the patients showed a moderate or significant improvement in glucose tolerance test results after taking the fruit juice before the glucose tolerance test (*51*). Unfortunately, the study was neither randomized nor blinded, and the patients' baseline characteristics were poorly described.

In a clinical study, 22 subjects (12 healthy volunteers and 10 patients with type II diabetes) were administered a powder of the fruit after which the serum cholesterol and glucose tolerance were assessed (*31*). Oral

administration of the powdered fruit to 10 patients with type II diabetes, at a dose of 2.0 g/day for 11 days, reduced blood glucose and total cholesterol levels by 10.02%. The reduction in blood glucose levels during the glucose tolerance test was 10.64–15.15% in the patients with diabetes, and was highly significant ($p < 0.001$).

The effects of the crude drug on fasting and post-prandial serum glucose levels (2 hours after oral administration of 75 g glucose) were studied in 100 subjects with moderate non-insulin dependent diabetes mellitus. Drinking the aqueous homogenized suspension of the vegetable pulp led to a significant reduction ($p < 0.001$) of both fasting and post-prandial serum glucose levels. This hypoglycaemic action was observed in 86 (86%) of the patients. Five patients (5%) showed lowering of fasting serum glucose only (*24*).

Immune stimulation

Cervical cancer patients have a decreased total white blood cell count, including that of natural killer cells. Natural killer cells, one type of lymphocyte, play a role in eliminating cancer cells by antibody-dependent cell-mediated cytotoxicity. A clinical study assessed the effect of the crude drug in cervical cancer patients undergoing normal treatment (radiotherapy). Subjects were divided into three groups:

— normal control (women aged 35–55 years, $n = 35$);
— patient control ($n = 30$); and
— patient treatment ($n = 30$).

The women in the patient control and patient treatment groups were cervical cancer patients (stage II or III) being treated with radiotherapy (without or with addition of the crude drug). Blood samples from women in the patient control and patient treatment groups were analysed for percentage of natural killer cells and concentration of P-glycoprotein. The results showed an increased percentage of natural killer cells in the patient control and patient treatment groups. The increase in both groups was significant ($p < 0.05$) when the percentage of natural killer cells from second and third blood samples (taken after radiation with or without addition of the crude drug for 45 and 90 days) was compared with that from the first blood sample (taken before treatment). The results for the women in the patient treatment group also showed a significant decrease of P-glycoprotein level ($p < 0.05$) in the second and third blood samples when compared with the first blood samples. There was no significant difference in the P-glycoprotein (P-gp) level between the first, second and third blood samples from the women in the patient control group. Ingestion of the crude drug did not affect numbers of natural killer cells, but it did affect the decrease of P-gp level on natural killer cell membrane (*52*).

Adverse reactions

The reported adverse effects of the crude drug include hypoglycaemic coma and convulsions in children, increases in γ-glutamyl transferase and alkaline phosphatase levels, and headaches (*53*).

Contraindications

Owing to potential abortifacient effects and possible teratogenicity (*54*, *55*), the seeds of the crude drug should not be taken during pregnancy.

Owing to reported adverse events such as severe hypoglycaemia and convulsions in children, the crude drug and its preparations should not be administered to children or taken during breastfeeding (*53*).

Warnings

Patients with liver disorders should seek advice from their health care professional before taking any crude drug preparation.

Precautions

Drug interactions

The fruit and preparations thereof may have additive effects when taken with other glucose-lowering agents; however these interactions need to be investigated (*53*).

Pregnancy: teratogenic effects

Momorcharins, isolated from the seeds of the crude drug have been shown to induce early and midterm abortions in mice and were teratogenic in cultured mouse embryos at the early organogenesis stage. Morphological abnormalities were observed in the head, trunk and limbs of the embryos (*54*, *55*).

The genotoxic potential of extracts of the crude drug was assessed in the *Salmonella typhimurium* microsome activation assay and the alkaline single-cell gel electrophoresis (COMET) assay. The extract did not produce a positive response in strains TA98 and TA100 with or without metabolic activation, but produced an increase above negative control values in the COMET assay (*56*).

Pregnancy: non-teratogenic effects

See Contraindications.

Breastfeeding mothers

See Contraindications.

Paediatric use
See Contraindications.

Other precautions
No information was found.

Dosage forms
Crude drug, extracts, fruit juice and tablets.

Posology
(Unless otherwise indicated)
Adult daily oral dose: 10–15 ml of fresh juice (*1*); 2–15 g of dried crude drug (*25*, *28*, *30*).

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Fructus Myrtilli

Definition
Fructus Myrtilli consists of the dried ripe fruits of *Vaccinium myrtillus* L. (Ericaceae) (*1*).

Synonyms
Vaccinium angelosums Dulac, *V. montanum* Salisb., *Myrtilis niger* Gilib. (*2*).

Selected vernacular names
Adara, aiges, airadech, airelle myrtille, aires, airolle myrtille, arándano, baceri mirtillo, baggiole, bagolo, Bickbeere, bilberry, bimbelas, blackberry, blaeberry, Blaubeere, Blaubessen, blue berry, blueberry, bog bilberry, brimbelle, burren myrtle, European blueberry, harilik, hei guo yue ju, Heidelbeere, Heidelbeerfruchten, huckleberry, maurettes, mirtillo nero, myrtille, petit myrtle, uva del boschi, uva orsina, waldbeere, whortleberry, wineberry (*2–4*).

Geographical distribution
Found in Europe and in the North American Rocky Mountains (*2*, *5*).

Description
Trailing shrub forming large colonies from creeping rhizomes, 10–60 cm in height; twigs green, glabrous, 3-angled. Leaves: deciduous, alternate, short petiolate; blade broadly elliptic to ovate, 6–18 mm wide, 10–30 mm long, apex acute to obtuse, base rounded; margin serrulate; bright green, lower surface sparsely glandular with prominent venation. Inflorescence: flowers solitary or paired in leaf axils, bracts 2. Flowers: perfect, radially symmetrical, 5-lobed; calyx lobes very short to almost absent; corolla pale green or white to pink, broadly urceolate to globose, 4–7 mm wide, 3–5 mm long, lobes very short and revolute; stamens 10, filaments glabrous, anthers awned, dehiscent by terminal pores; ovary inferior, style usually included. Fruit: berry, oblate-globose, 5–9 mm diameter, blue to black, rarely glaucous, many-seeded. Chromosome number: 2x = 2n = 24 (*2*).

Plant material of interest: dried ripe fruit

General appearance

The dried berry is a dark blue, subglobular, shrunken berry, about 5 mm in diameter, with a scar at the lower end and coarsely wrinkled exocarp. The pedicel may be attached or detached, and often the remains of the style, nectar disc, and calyx are persistent at the apex of the berry, with the calyx appearing as a circular fold. The mesocarp is purple. There are 4–5 locules, each containing many seeds; each seed is approximately 1 mm long with a yellowish brown dimpled surface (*1*).

Organoleptic properties

Odour: no characteristic odour; taste: sweet and slightly bitter (*1*).

Microscopic characteristics

The exocarp consists of polygonal, rectangular or quadratic cells with slightly pitted tangential walls. Groups of 2–4 cells occur, each group surrounded by a thick wall, while within the groups the walls are considerably thinner. The mesocarp consists of large parenchymatous cells with scattered solitary sclereids and vascular bundles containing vessels with spiral or helical secondary wall thickenings. The endocarp is composed largely of groups of sclereids similar to those in the mesocarp and having an elongated or nearly quadratic shape. The outer layer of the testa consists of elongated, heavily thickened and pitted sclereids. In cross-section these cells have U-shaped secondary walls with the unthickened side occurring on the outer tangential wall. The endosperm cells are thin-walled and contain droplets of fixed oil. Calcium oxalate crystals may occur occasionally in all tissues (*2*).

Powdered plant material

Violet-pink sclereids from the endocarp and the mesocarp and testa, usually aggregated, with thick, channelled walls; reddish brown fragments of the epicarp consisting of polygonal cells with moderately thickened walls; brownish yellow fragments of the outer seed testa made up of elongated cells with U-shaped thickened walls; calcium oxalate crystals of various sizes as clusters and prisms. Also present are parenchyma cells, vascular bundles and oil droplets (*1*).

General identity tests

Macroscopic and microscopic examinations, thin-layer chromatography (*1*, *2*), and high-performance liquid chromatography (*6*, *7*).

Purity tests

Microbiological

Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*8*).

Foreign organic matter

Plant material complies with the test for foreign matter (*1*).

Total ash

Not more than 5% (*1*).

Acid-insoluble ash

To be established in accordance with national requirements.

Water-soluble extractive

To be established in accordance with national requirements.

Alcohol-soluble extractive

To be established in accordance with national requirements.

Loss on drying

Not more than 12% (*1*).

Pesticide residues

The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*1*). For other pesticides, see the *European pharmacopoeia* (*1*) and the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*8*) and pesticide residues (*9*).

Heavy metals

For maximum limits and analysis of heavy metals, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*8*).

Radioactive residues

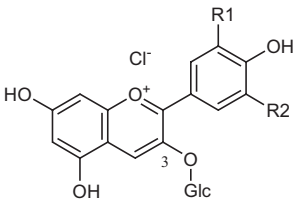
Where applicable, consult the WHO guidelines on assessing quality of herbal medicines with reference to contaminants and residues (*8*).

Chemical assays

Not less than 1.0% of tannins, expressed as pyrogallol (*1*).
Not less than 0.2% of anthocyanins, expressed as cyanidin-3-glucoside chloride (*2*).

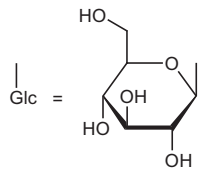
Major chemical constituents

Besides the normal organic acids (3–7%; e.g. citric acid and malic acid), phenolic acids and other phenols, the fruits contain up to 10% tannins (mostly catechol tannins). The major characteristic and biologically active constituents are flavonoids and anthocyanins. Examples of flavonol-*O*-glycosides in fresh bilberry juice and fruit include quercetin-3-rhamnoside (quercitrin), quercetin-3-glucoside (isoquercitrin), quercetin-3-galactoside (hyperoside), and kaempferol-3-glucoside (astragalin). More than 15 anthocyanins (totalling approximately 0.5%) have been identified as 3-arabinosides, 3-glucosides and 3-galactosides of five anthocyanidins: cyanidin, delphinidin, malvidin, peonidin and petunidin, with cyanidin and delphinidin glycosides accounting for 64% of the total anthocyanins (*2*, *3*, *5*, *10*). Structures of the 3-glucosides of cyanidin, delphinidin, malvidin, peonidin and petunidin are presented below.



Cyanidin R1 = OH R2 = H
Delphinidin R1 = R2 = OH
Malvidin R1 = R2 = OCH_3
Peonidin R1 = OCH_3 R2 = H
Petunidin R1 = OH R2 = OCH_3

3-monoglucosides



β-D-glucopyranosyl

Medicinal uses

Uses supported by clinical data

Oral use for the symptomatic treatment of dysmenorrhoea associated with premenstrual syndrome (*11*), circulatory disorders in patients with capillary leakage or peripheral vascular insufficiency (*12–16*) and ophthalmic disorders (*17–19*).

Uses described in pharmacopoeias and well established documents

Oral use for the treatment of acute diarrhoea and local irritation or inflammation of the mucous membranes of the mouth and throat (*20*).

Uses described in traditional medicine

Treatment of capillary fragility, diarrhoea, eye disorders, haemorrhoids, intestinal disorders, skin disorders and venous insufficiency. Also used as a diuretic (*2*, *3*).

Pharmacology

Experimental pharmacology

Anti-inflammatory activity

An extract of the fruit containing 25% anthocyanidins had vasoprotective and anti-oedema (anti-inflammatory) activities in experimental animals (*21*). In rabbits, chloroform-induced skin capillary permeability was reduced after intraperitoneal administration of the extract at a dose of 25.0–100.0 mg/kg body weight (bw) or intragastric administration at a dose of 200.0–400.0 mg/kg bw of anthocyanosides. The anti-inflammatory effect of the extract lasted longer than that of the comparison treatments rutin or mepyramine. Intragastric administration of the anthocyanins to rats, at a dose of 25.0 to 100.0 mg/kg bw was effective treatment both in the skin capillary permeability test and on vascular resistance of animals fed a vitamin P-deficient diet. In both the animal models (rats and rabbits), the anthocyanins were twice as effective as the flavonoid, rutin. Furthermore, intragastric administration of the anthocyanins (dose not stated) inhibited carrageenan-induced paw oedema in rats after intravenous injection or topical application (*21*).

Antioxidant activity

An aqueous extract of the fruit inhibited copper-induced oxidation of human low-density lipoprotein in vitro. Results were obtained by measurement of oxidative resistance as determined by the lag-phase preceding conjugated diene formation; quantification of the amount of lipoperoxides, as well as thiobarbituric acid-reactive substances generated, and measurement of the modification in the net negative electrical charge of the lipoproteins, over a 7-hour time course experiment. Trace amounts of the extract, at concentrations of 15 to 20 µg/ml induced statistically significant changes in the oxidation behaviour of low-density lipoprotein, which included: prolongation of the lag-phase of conjugated diene production ($p < 0.01$); reduction in the formation of lipoperoxides and of thiobarbituric acid-reactive substances up to 7 hours and especially between 1 and 5 hours ($p < 0.01$); and inhibition of modification in the net negative charge of low-density lipoprotein. These results demonstrate that the extract exerts potent protective action on low-density lipoprotein particles during in vitro copper-mediated oxidation (*22*). An anthocyanin-containing extract of the fruit inhibited lipid peroxidation and hydroxyl radical formation with a median inhibitory concentration of 50.3 µg/ml in rat liver microsomes ($p < 0.01$). The extract also exhibited superoxide scavenging activity (25.0 µg/ml, $p < 0.01$) (*23*). A study to compare the concentration of phenolics and anthocyanins and the anti-

oxidant activity of the berries of various *Vaccinium* cultivars (including the crude drug) was performed (*24*). Total antioxidant capacity, measured as oxygen radical absorbance capacity, ranged from 13.9 to 45.9 µmol/g of fresh berries in the extract. Bilberry and the lowbush blueberries from Nova Scotia had the highest antioxidant activity (44.6 and 45.9 µmol/g, respectively).

Radical scavenging properties of an extract or tea of the fruit containing anthocyanins was tested on the 1,1-diphenyl-2-picrylhydryl radical using electron spin resonance spectroscopy. Both the extract and the tea were effective 1,1-diphenyl-2-picrylhydryl radical scavengers. No direct correlation was found between the scavenging activity and the content of anthocyanosides and flavonoids in the extract, suggesting that the catechins and ascorbic acid may also play a role (*25*).

Pharmacokinetics

Administration to rats of a fruit extract containing anthocyanins, at a dose of 20–40 mg/kg bw (intravenous) or 25.0 mg/kg bw (intraperitoneal) underwent rapid body distribution. Elimination occurred primarily in the urine and bile, following a three-compartment pharmacokinetic model. After 4 hours, 20% of the dose was eliminated in the urine, regardless of route of administration, while at 24 hours, 15% and 18% of the dose was eliminated in the bile after intravenous and intraperitoneal administration, respectively. The anthocyanins possessed a greater affinity for some tissues, namely kidneys (79.0 µg/g tissue) and skin (27.4 µg/g tissue) than for plasma (19.0 µg/g) (*26*).

The pharmacokinetics were assessed after intravenous administration of anthocyanosides (20.0–40.0 mg/kg bw) or oral administration to rats (400.0 mg/kg bw). The results of the intravenous dose were the same as those reported by Lietti and Forni (*26*). After a single oral administration, the plasma concentrations of anthocyanins reached a peak after 15 minutes and then rapidly declined within 2 hours. The extent of cumulative urinary and biliary elimination, together with the gastrointestinal recovery, demonstrated an absorption rate of approximately 5%. No hepatic first-pass effect was observed. Despite the modest gastrointestinal absorption and the low absolute bioavailability (1.2% of the administered dose), the peak levels in plasma (2.0–3.0 µg/ml) measured after the oral treatment are in the range of biological activity reported for these substances (*27*).

Vascular permeability

The anthocyanins contained in the fruit are thought to have "vitamin P" activity in that they increase the levels of intracellular vitamin C and de-

crease capillary permeability and fragility (*26*). In one study, the initial phase of renal hypertension induced by ligature of the abdominal aorta was accompanied by a transient increase in vascular permeability. This increase in permeability is higher in the skin and in the aorta wall than in the blood vessels of the brain. Treatment of rats with anthocyanins from the crude drug (dose not stated) for 12 days prior to the induction of hypertension kept the blood–brain barrier permeability normal and limited the increase in vascular permeability in the skin and the aorta wall (*28*). In cholesterol-fed rabbits, administration of anthocyanidins from the crude drug (dose not stated) did not modify the serum cholesterol levels, but decreased the proliferation of the intima; the extracellular matrix production; the deposition of calcium and lipid in the aorta; and decreased the DNA and lipid contents. Alteration in the biochemical composition of the isolated brain microvessels was also diminished. The suggested mechanism of action may be through an interaction of the anthocyanins with collagen, increasing the cross-linking and thus diminishing the permeability of small, as well as of large, blood vessels (*29*).

The effects of anthocyanosides from the fruit on capillary filtration in diabetic rats were assessed. Rats with streptozotocin-induced diabetes were randomly allocated to one of three groups to receive either *Ginkgo biloba* (group A), the crude drug (group B) or no treatment (group C). The isotopic test of capillary filtration consisted of intravenously injecting ^{99}mtechnetium-labelled albumin, inducing venous compression on a hindquarter, and measuring radioactivity externally on the limb before, during and after removal of venous compression. Interstitial albumin retention and the ratio of the amplitudes of the low- and high-frequency peaks (LF/HF ratio), an index of lymphatic function obtained by the fast Fourier transform of the last part of the radioactivity curve, were calculated. In streptozotocin-treated animals, the isotopic test was performed at a mean age of 97 days (time 1) and after 6 weeks (time 2) and 12 weeks (time 3) of treatment (6 and 12 weeks after time 1). At time 1, albumin retention was significantly higher in the three groups of diabetic rats than in the control rats, with no significant difference between these groups. In group B, albumin retention decreased significantly at times 2 and 3 ($p = 0.015$). In group C, albumin retention increased significantly from time 1 to time 3 ($p < 0.0005$). In group A, albumin retention increased slightly (not statistically significantly) between time 1 and time 3. In groups A and C, the LF/HF ratio significantly increased with time ($p < 0.0005$) and the levels at time 3 were significantly higher than those in control rats ($p < 0.0001$). In group B, the LF/HF ratio remained unchanged from time 1 to time 3 and was similar to the values found in the control

rats. The study demonstrated that anthocyanins from the crude drug are effective in preventing the increase in capillary filtration and the failure of lymphatic uptake of interstitial albumin in diabetic animals (*30*).

The effects of an extract of the crude drug containing anthocyanosides on ischaemia reperfusion injury were investigated in the hamster cheek pouch microcirculation. Ischaemia was induced by clamping the cheek pouch for 30 min followed by 30 min of reperfusion. The microvasculature was visualized by a fluorescence technique. The extract was administered orally at a dose of 100.0 mg/kg bw for 2 and 4 weeks. The outcomes measured were the number of leukocytes adhering to venular vessel walls, the perfused capillary length, the increase in permeability and the changes in arteriolar diameter. Ischaemia and reperfusion were associated with an increased number of leukocytes sticking to venules, a decreased number of perfused capillaries and increased permeability. Administration of the extract decreased the number of leukocytes sticking to the venular wall and preserved the capillary perfusion; the increase in permeability was significantly reduced after reperfusion. Administration of the extract preserved the arteriolar tone and induced the appearance of rhythmic changes in the diameter of arterioles, indicating that the extract reduced microvascular impairments due to ischaemia reperfusion injury, with preservation of endothelium, attenuation of leukocyte adhesion and improvement of capillary perfusion (*31*).

The effects of anthocyanins from the crude drug on arteriolar vasomotion were assessed in cheek pouch microcirculation of anaesthetized hamsters and in skeletal muscle microvasculature of unanaesthetized hamster skin fold window preparation. Intravenously injected anthocyanins induced vasomotion in cheek pouch arterioles and terminal arterioles with higher frequency in smaller vessels. In the arteriolar networks of skeletal muscle, anthocyanins increased vasomotion frequency and amplitude in all orders of vessel. The results indicate that anthocyanins are effective in promoting and enhancing changes in arteriolar rhythmic diameter that play a role in the redistribution of microvascular blood flow and interstitial fluid formation (*32*).

Clinical pharmacology

Cataracts and glaucoma

In one randomized, double-blind, placebo-controlled study involving 50 patients with mild senile cortical cataracts, a standardized extract of the crude drug containing 25% anthocyanosides was administered at a dose of 180.0 mg twice daily together with vitamin E, in the form of dl-tocopheryl acetate (100 mg twice daily) or placebo for 4 months. The treatment

retarded the progression of cataracts in 97% ($p < 0.05$) of the subjects ($n = 25$) compared to 76% in the control group ($n = 25$) (*17*). However, several studies have also shown that administration of vitamin E alone reduces the incidence of cataracts (*18*).

Extracts of the crude drug have also been tested in the treatment of glaucoma. In one small pilot study, eight patients with glaucoma were given a single dose of an extract of the crude drug containing 200.0 mg of anthocyanosides. Electroretinography showed improvements in all patients; however no further details are available (*18*, *33*).

Diabetic retinopathy

In one report, oral administration of bilberry anthocyanins, at a dose of 600.0 mg/day for 6 months, to 32 patients with diabetes reduced the number of capillaries with lesions from 34% before treatment to 14% after treatment (*2*). In another investigation, 31 patients with various types of retinopathy were treated with an extract of the crude drug to determine the effect of anthocyanins on the retinal vessels. In patients with diabetic retinopathy, a reduction in permeability and tendency to haemorrhage was observed (*34*). In a randomized, double-blind, placebo-controlled study the effect of the crude drug was investigated in 40 patients with diabetic and/or hypertensive retinopathy. Patients were divided into two equal groups and either treated with a crude drug extract equivalent to 115 mg anthocyanosides daily, or a placebo, for 1 month. Retinopathy in the 20 patients receiving placebo remained unchanged and these patients were treated for a further 30 days. At the end of treatment (30 or 60 days), detectable retinal abnormalities (seen in 13/20 patients in the treatment group initially) were reduced in 10 patients and, in three, symptoms were unchanged. In the placebo group, retinal abnormalities (seen in 15/20 patients initially) were unchanged after 30 days, but when they were given active treatment for a further 30 days, 79% of patients improved (*19*).

Myopia

An extract of the crude drug containing 160 mg anthocyanins was investigated for its effect on myopia in 26 patients. Improvement of scotopic function was observed in all patients, but the effects were only statistically significant in subjects with slight myopia (≤ 6 diopters) (b2 wave, $p < 0.01$). In subjects with medium myopia, photopic function was significantly improved (critical central fusion frequency, $p < 0.005$; b1 wave, $p < 0.01$) (*2*). The effect of a preparation containing anthocyanosides and vitamin E on refraction, visual acuity and eye fundus was assessed in 36 patients with progressive myopia. After an observation period of 14.5 months an average increase of myopia by 0.53 diopters per eye was

demonstrated. The final examination of 29 patients showed a stabilization of the fundus alterations, as well as a stable, or an improved visual acuity. In seven patients, a moderate deterioration of the partial or overall medical condition occurred (*35*).

Night vision improvement

The effect of the crude drug on the enhancement of night vision was first investigated by researchers in studies of Royal Air Force pilots during the Second World War. In these case-reports, the investigators suggested that there were improvements in night vision less than 24 hours after ingesting an unknown quantity of bilberry jam (*36*). Administration of the bilberry jam resulted in improved nighttime visual acuity, faster adjustment to darkness and faster restoration of visual acuity after exposure to glare (*37*, *38*). Later studies supported these observations (*36*, *39*, *40*). Two studies showed that the administration of four tablets of an unspecified extract of the crude drug (100 mg per tablet) increased the light sensitivity threshold (*36*). However, these results have not been duplicated in controlled clinical trials. In one study, the ability of anthocyanosides in a single oral dose to improve night vision in normal individuals was evaluated during three night vision tests: full-field scotopic retinal threshold, dark adaptation rate and mesopic contrast sensitivity (*41*). The study, a double-blind, placebo-controlled, cross-over study, involved 16 young normal volunteers who were randomly assigned to one of four different regimens of single oral administrations of 12, 24 and 36 mg of anthocyanosides or a placebo, with a 2-week washout period between doses. Scotopic retinal threshold, dark adaptation rate and mesopic contrast sensitivity were measured immediately before, and 4, 8 and 24 hours after treatment. No significant effect of the treatment was found on any of the three night vision tests during the 24 hours following administration. The study concluded that single oral administration of 12–36 mg of anthocyanosides lacked any significant effect on night vision tests relevant to the military (*41*).

In a controlled study, the ability of multiple oral doses of anthocyanosides to improve night vision in normal individuals was assessed. The effect was tested in the three night vision tests: scotopic retinal threshold, dark adaptation rate and mesopic contrast sensitivity. This double-blind, placebo-controlled, cross-over study involved 18 young normal volunteers who were randomly assigned to one of three different regimens of oral administrations of 12.0 and 24.0 mg/day anthocyanosides or a placebo, given twice daily for 4 days. A 2-week washout period was allowed between each 4-day treatment period. Scotopic retinal threshold, dark adaptation rate and mesopic contrast sensitivity tests were done 1 day before treatment and on days 1, 2, 3 and 4 during the treatment period.

Again, no significant effect on any of the three above-mentioned night vision tests was found (*42*).

Another double-blind, placebo-controlled, cross-over study conducted on US Navy SEALs personnel investigated the effect of bilberry on night visual acuity and night contrast sensitivity. The test subjects were young men with good vision; eight received a placebo and seven received active capsules containing 160 mg of bilberry extract (25%) for 3 weeks. After the 3-week treatment period, a 1-month washout period was used to allow any effect of bilberry on night vision to dissipate. In the second 3-week treatment period, the eight subjects who first received the placebo were given active capsules, and the seven who first received active capsules were given the placebo. Night visual acuity and night contrast sensitivity were again tested throughout the 3-month experiment. The results of this investigation showed no difference in night visual acuity during any of the measurement periods when examining the average night visual acuity or the last night visual acuity measurement during active and placebo treatments. In addition, there was no difference in night contrast sensitivity during any of the measurement periods when examining the average night contrast sensitivity or the last night contrast sensitivity measurement during active and placebo treatments (*36*).

Premenstrual syndrome and dysmenorrhoea

In a randomized, double-blind, placebo-controlled clinical trial the efficacy of a fruit extract containing anthocyanosides on the symptoms of dysmenorrhoea associated with premenstrual syndrome was assessed. Women with primary dysmenorrhoea received either the extract corresponding to 115 mg anthocyanosides per day or a placebo for 5 days beginning 3 days before menstruation for two consecutive cycles. A significant improvement in pelvic/lumbosacral pain ($p < 0.01$), mammary tension ($p < 0.01$), nausea ($p < 0.01$), and heaviness of the lower limbs ($p < 0.01$) was reported as compared with the baseline (*11*).

Venous insufficiency and varicose veins

The ability of an extract of the crude drug containing anthocyanins to improve symptoms associated with varicosities and telangiectases was assessed. Twenty-seven patients with varices, varicosities and telangiectases were treated orally with 4–6 tablets, equivalent to 100–150 mg anthocyanins daily for 10–15 days per month for 2 months. An improvement in a variety of symptoms was observed including a reduction in bruising (*12*). In one study in humans, oral administration of a crude drug extract containing anthocyanins (dose not stated) to patients with varicose veins and ulcerative dermatitis reduced capillary leakage (*16*). The biochemical and

histochemical data suggested that the anthocyanins protect the capillary walls by a mechanism that involves increasing the endothelium barrier-effect through stabilization of the membrane phospholipids and by increasing the biosynthetic processes of the acid mucopolysaccharides of the connective ground substance, by restoring the altered mucopolysaccharidic pericapillary sheet. A marked increase in newly formed capillaries and collagen fibrils induced by the anthocyanins was also observed (*16*). In another study, 47 patients with varicose veins were treated with a commercial extract of the crude drug at a dose of 480 mg/day for 30 days (*14*). Significant improvements in microcirculation, oedema, feelings of heaviness, parasthesia, pain and skin dystrophy were observed ($p < 0.01$). A significant reduction in oedema was observed by day 15 ($p < 0.01$). In another similar study, 15 patients with polyneuritis due to peripheral vascular insufficiency were given 480 mg/day of the extract and a significant improvement in microcirculation was noted (*43*). Furthermore, a review of uncontrolled trials from 1979 to 1985 involving a total of 568 patients with venous insufficiency of the lower limbs showed that an anthocyanin-containing extract of the crude drug was effective in rapidly decreasing symptoms and improving both venous microcirculation and lymph drainage (*13*).

Pharmacokinetics and toxicology

A randomized parallel dietary intervention study measured the serum quercetin concentrations of 40 healthy subjects who were consuming fruit (including the crude drug) or normal Finnish diets. Twenty subjects consumed 100 g/day of berries (blackcurrants, lingon berries and bilberries) for 8 weeks. Twenty subjects consuming their normal diets served as controls. Fasting blood samples were obtained 2 weeks prior to the study, at baseline, and at 2, 4 and 8 weeks. Intake of quercetin was assessed from 3-day food records collected at baseline and at 8 weeks. The serum quercetin concentrations were significantly higher in the subjects consuming berries than in subjects in the control group ($p = 0.039$; analysis of variance with repeated measures). During the period of berry consumption the mean serum concentrations of quercetin ranged between 21.4 and 25.3 µg/l in the group consuming the berries; this was 32–51% higher than in the control group. According to the 3-day food records, there was no difference in quercetin intake at baseline, but at 8 weeks the intake was 12.3 mg/day (mean) in the group consuming the berries and 5.8 ± 0.6 mg/day in the control group ($p = 0.001$) (*44*).

Long-term oral administration to humans of doses equivalent to 180 mg/kg anthocyanins per day for 6 months produced no toxic effects (*2*).

Adverse reactions
No information was found.

Contraindications
No information was found.

Warnings
If diarrhoea persists for more than 3–4 days, or is associated with abdominal pain or rectal bleeding, consult a health care professional.

Precautions
Drug interactions
In one ex vivo study, oral administration of an extract of the fruit containing anthocyanins inhibited platelet aggregation when given at doses of 480 mg daily for 30–60 days (*45*).

Therefore, fruit extracts have antiplatelet aggregating properties and very high doses should be used cautiously in patients with haemorrhagic disorders and those taking anticoagulant or antiplatelet drugs.

Carcinogenesis, mutagenesis, impairment of fertility
No mutagenicity or carcinogenicity has been reported.

Pregnancy: teratogenic effects
No teratogenic effects were observed in animals treated with 3–5 times the human dose (*2*).

Other precautions
No information was found.

Dosage forms
Crude drug, extracts, tablets and capsules.

Posology
(Unless otherwise indicated)
Internal: daily dosage of crude drug 20–60 g (*2*). Extracts: 80–160 mg of extract standardized to 25% anthocyanosides (three times daily). The dose of anthocyanosides is 20–40 mg three times daily (*20*). External: 10% decoction; equivalent preparations (*20*).

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Radix Panacis Quinquefolii

Definition
Radix Panacis Quinquefolii consists of the dried roots of *Panax quinquefolius* L. (Araliaceae) (*1–3*).

Synonyms
Aralia quinquefolia Dec. & Planch., *Ginseng quinquefolium* Wool, *Panax americanum* Raf. (*4*).

Selected vernacular names
Ameerika zensen, American ginseng, American white ginseng, Amerika ninjin, Amerikanischer Ginseng, Amerikanne Ginseng, Canadian ginseng, Canadian white ginseng, Chinese seng, five fingers, garantogen, ginseng americano, ginseng d'Amerique, hsi-yang-shen, hua-ch'i-seng, hua-ch'i-senh, little Indian, man root, matc'etasa, ninsin, North American ginseng, seng, seiyou ninjin, shang, wenane, wild American ginseng, xi yang shen, xi-yang-shen (*5*, *6*).

Geographical distribution
Native to Canada and the United States, and cultivated in France and northern China (*7*).

Description
Herb, up to more than 1 m high; rootstock fusiform, up to 2 cm in diameter; stem straight, slender, subterete, often striate; leaves 3 or 4, 5 (3–7)-foliolate, petiole slender, up to 10 (rarely to 15) cm long, leaflets thin, elliptic to obovate, up to 16 cm long and 8.5 cm broad (the basal ones smaller, often ovate), acute to rounded at the base, acuminate at the apex, conspicuously and often doubly serrate, the teeth deltoid, acute, the petiolules up to 4.5 cm long, the principal veins slightly raised on both surfaces, lateral veins 5–9, ascending; peduncle slender, straight, up to 10 (rarely to 30) cm long, the bractlets deltoid to lanceolate, acute, 2–5 mm long; pedicels 15–40 per umbel, up to 12 mm long, swollen distally; calyx

carnose, cupuliform, at anthesis about 2 mm long and 2 mm in diameter, the lobes deltoid, acute, about 0.5 mm long; petals greenish-white, membranous, slightly granular-papillose distally, oblong, about 1.5 mm long and 1 mm broad, subacute and slightly incurved at the apex; filaments carnose, narrowed distally, 1–1.5 mm long, the anthers oblong, about 1 mm long, obtuse at both ends; summit of the ovary flattened or concave, styles 2, carnose, slightly curved, 1–1.5 mm long; locules 2; fruit laterally flattened, transversely oblong, up to 7 mm long and 10 mm broad, longitudinally sulcate, the wall at length dry, seeds 2, oblong, 4–5 mm long, 3–4 mm broad (*4*).

Plant material of interest: dried roots

General appearance

Fusiform, cylindrical or conical, 1–12 cm, sometimes up to 20 cm in length, and up to 2.5 cm in diameter at the crown, with one or more stem scars. Externally pale yellow to golden, exhibiting transverse striations and linear lenticels, and showing fine and dense longitudinal wrinkles, and rootlet scars. If stem base is present, then scales thin and perishing (differs from *P. ginseng*, in which scales at base of stem are fleshy and persistent). The middle and lower part of the main root with 1 or more lateral roots, mostly broken off. Sometimes, the upper part with rhizome showing distinct annulations, rounded or semi-rounded stem scars, and bearing adventitious roots, or broken off. Texture heavy and hard, not easily broken, fracture short, fractured surface is white to ivory, with distinct aromatic odour and rings of secretory canals present in secondary phloem, slightly starchy, bark exhibiting yellowish brown dotted resin canals, cambium ring brownish yellow, wood exhibiting less distinct radiate striations (*1*, *2*).

Organoleptic properties

Odour: slight and characteristic; taste: slightly bitter and sweet (*1*, *2*).

Microscopic characteristics

In contrast to *Panax ginseng*, transverse sections of *P. quinquefolius* show abundant large, thin-walled cork parenchyma cells. Secondary phloem is characterized by conspicuous air lacunae; abundant starch-containing storage parenchyma; few sieve elements, found in small groupings; and rings of schizogenous secretory canal lined with 6–8 epithelial cells that lack starch. Xylem is characterized by abundant starch-containing storage parenchyma and a few tracheary elements, composed of non-lignified tracheids and slightly lignified spiral or reticulated vessels lacking secretory canals and found in isolation or in small groupings. Druse crystals

are sometimes present within vascular parenchyma cells. Diarch or triarch primary xylem is in the centre of root starch grains, simple or 2–5-compound, range from 5–6 µm in diameter (*1*, *8*).

Powdered plant material

Pale yellowish brown powder with a slightly aromatic odour. Shows cork cells; yellowish secretory canals and yellowish brown secretory substances in and outside the canal; fragments of parenchyma cells contain starch grains, simple or 2–5-compound, range from 5–6 µm in diameter; tracheids with spiral or reticulate thickening rings; irregular shaped bast fibres, lumen smooth, 200–1200 µm in length; rosettes of calcium oxalate 30–33 µm in diameter (*1*, *8*).

General identity tests

Macroscopic (*1*, *2*) and microscopic examinations (*1*, *8*); DNA analysis (*9*); and thin-layer chromatography (*1*, *2*, *10*) and high-performance liquid chromatography (*1*, *2*, *11*, *12*) for the presence of 24(*R*)-pseudoginsenoside F11 and the absence of ginsenoside Rf.

Purity tests

Microbiological

Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*13*).

Foreign organic matter

Not more than 2.0% (*1*).

Total ash

Not more than 8.0% (*1*).

Acid-insoluble ash

Not more than 1% (*2*).

Water-soluble extractive

To be established in accordance with national requirements.

Alcohol-soluble extractive

Not less than 30% (*2*).

Loss on drying

Not more than 10.0% (*1*).

Pesticide residues

The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*14*). For other pesticides, see the *European pharmacopoeia* (*14*) and the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*13*) and pesticide residues (*15*).

Heavy metals

For maximum limits and analysis of heavy metals, consult the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*13*).

Radioactive residues

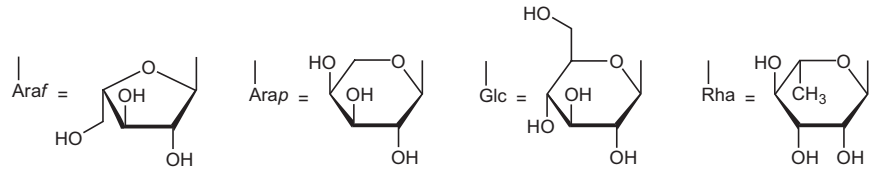
Where applicable, consult the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*13*).

Chemical assays

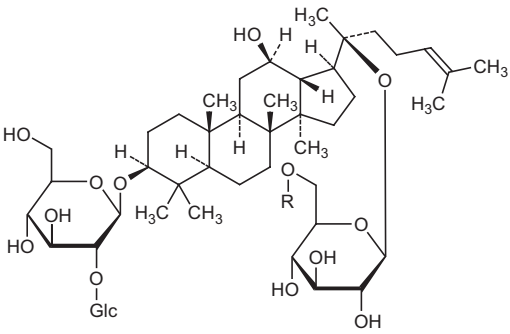
Not less than 2% total ginsenosides of Rg1, Re and Rb1 (*2*).

Major chemical constituents

Major constituents of the root are the damamrane triterpene saponins collectively known as ginsenosides. As in the case of *Panax ginseng*, the ginsenosides of *P. quinquefolius* are derivatives of protopanaxadiol or protopanaxatriol, with the majority of these compounds (e.g. ginsenosides Rb1, Rb2, Rc, Rd, Re, Rg1 and Ro) being common to both species. However, there are quantitative and qualitative differences. The total ginsenoside content of *Panax quinquefolius* is higher than that of *Panax ginseng* whereas the ginsenosides Rf and Rg2 do not occur in *P. quinquefolius*. On the other hand, 24(*R*)-pseudoginsenoside F11 is found in *P. quinquefolius*, but not in *P. ginseng*. In cultivated *P. quinquefolius*, however, the dominant ginsenosides are malonyl (m)-Rbl, Rb1 and Re with the percentages of m-Rb1 and Rb1 being almost identical. (Rg1 levels and total ginsenosides are much higher in wild than in cultivated *P. quinquefolius*.) (*6*). Furthermore, the combined amount of Rbl and m-Rb1 often exceeds half of the total ginsenoside content with the total malonyl ginsenoside (m-Rb1, m-Rb2, m-Rc and m-Rd) content being approximately 40% (*5*, *6*). In a study of wild American ginseng, total ginsenosides range from 1–16%, with the majority being in the range of 4–5% (*16*). Polysaccharides of biological significance include quinquefolans A, B and C (*17*). The structures of ginsenosides Rb1, Rb2, Rc, Rd, Re, Rg1 and pseudoginsenoside F11 are presented below.



α-L-arabinofuranosyl α-L-arabinopyranosyl β-D-glucopyranosyl α-L-rhamnopyranosyl

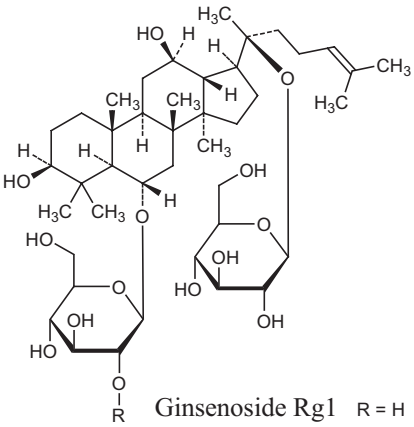


Ginsenoside Rb1 R = Glc

Ginsenoside Rb2 R = Arap

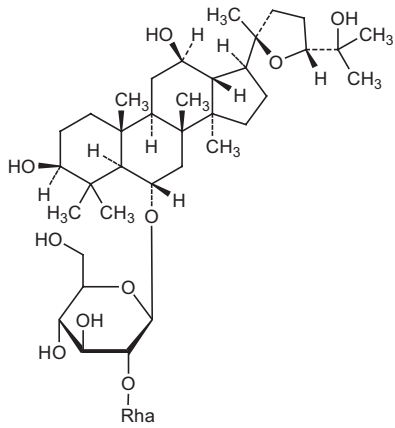
Ginsenoside Rc R = Araf

Ginsenoside Rd R = H



Ginsenoside Rg1 R = H

Ginsenoside Re R = Rha



Pseudoginsenoside F11

Medicinal uses

Uses supported by clinical data

None.

Uses described in pharmacopoeias and well established documents

While some preliminary studies have indicated that the root may be useful as an adjunct for the management of postprandial hyperglycaemia in subjects with type 2 diabetes mellitus (*18–22*), the numbers of subjects participating in the studies was too small to enable any solid conclusions

to be drawn. Further randomized controlled clinical trials with larger populations of patients with diabetes and better methodology are needed before therapeutic recommendations can be made.

Uses described in traditional medicine

Used orally as a diuretic, digestive, tonic and a stimulant (*5*, *6*, *23*). Used to enhance stress resistance, and to treat cough, loss of appetite, colic, vomiting, insomnia, neuralgia, rheumatism and headaches (*5*, *24*).

Experimental pharmacology

Antihyperglycaemic activity

Intraperitoneal administration of an aqueous extract of the root exhibited significant hypoglycaemic activity in mice with alloxan-induced hyperglycaemia when administered at a dose of 10 g/kg body weight (bw) ($p < 0.01$). Activity-guided fractionation of the extract led to isolation of three glycans, the quinquefolans A, B and C, which displayed significant hypoglycaemic activity ($p < 0.01$, $p < 0.05$, $p < 0.01$, respectively) in normal mice and in mice with alloxan-induced hyperglycaemia at a dose of 10, 100 and 10 mg/kg bw, of quinquefolans A, B and C, respectively (*17*).

Antioxidant activity

The antioxidant activity of a root extract containing 8% ginsenosides was assessed in the metal chelation, affinity to scavenge 1,1-diphenyl-2-picrylhydrazyl stable free radical, and peroxyl and hydroxyl free radical assays. Dissociation constants (Kd) for the extract to bind transition metals were of the order of $Fe^{2+} > Cu^{2+} > Fe^{3+}$ and corresponded to the affinity to inhibit metal-induced lipid peroxidation. In a metal-free linoleic acid emulsion, the extract exhibited a significant concentration-dependent (0.01–10 mg/ml) mitigation of lipid oxidation as assessed by the ammonium thiocyanate method ($p \leq 0.05$). The extract also showed strong 1,1-diphenyl-2-picrylhydrazyl radical scavenging activity up to a concentration of 1.6 mg/ml ($r^2 = 0.996$). The extract also inhibited the non-site-specific deoxynucleic acid (DNA) strand breakage caused by Fenton agents, and suppressed the Fenton-induced oxidation of a 66 Kd soluble protein obtained from mouse brain over a concentration range of 2.0–40.0 mg/ml (*25*). The free radical scavenging capacity and antioxidant effects of the crude drug appear to be related to the concentration of ginsenosides Rb1 and Rb2 in the plant (*26*).

The effect of saponins, isolated from the root, on the oxidation of low-density lipoprotein (LDL) was measured. The potential of the saponins for reducing LDL oxidation as well as limiting the ability of oxidized-LDL

(Ox-LDL) to impair endothelium-dependent relaxation in rat aortic rings was specifically assessed. Native LDL (0.2 or 0.3 mg/ml) was incubated with the saponins (0.25–1.0 mg/ml), with vitamin C (50 µM) as a control. Oxidation was initiated with copper sulfate at 37 °C for 0–24 h. The results demonstrate that the saponins reduced lipid peroxide levels concentration-dependently and also reduced alterations in relative electrophoretic mobility of Ox-LDL in a similar manner. Furthermore, measurement of content of phospholipid fractions indicated that the saponins reduced the conversion of phosphatidylcholine to lysophosphatidylcholine in Ox-LDL (*27*). Further investigations indicated that lower concentrations of saponins (1.0–100.0 µg/ml) in combination with vitamin C (1.0–10.0 µM) significantly enhanced the protective effects of the saponins. Pretreatment with the saponins (1.0–100.0 µg/ml) resulted in concentration-dependent inhibition of LDL oxidation and prolongation of lag time (*28*).

Antithrombotic activity

The effects of a root extract on thrombin-induced endothelin release were investigated using cultured human umbilical vein endothelial cells. Endothelial cells were pretreated with 1, 10 and 100.0 µg/ml of the extract and then incubated for 4 and 24 h with thrombin. The results demonstrated that the concentration of endothelin was significantly decreased in a concentration-dependent, time-related manner (at 4 h, 50% inhibitory concentration (IC_{50}) = 5.1 µg/ml; at 24 h, IC_{50} = 6.2 µg/ml). Furthermore, pretreatment of cultured endothelial cells with NG-nitro-L-arginine, a nitric oxide synthetase inhibitor, significantly ($p < 0.05$) inhibited thrombin-induced endothelin release by the extract indicating that the effect was partly due to release of nitric oxide (*29*).

Hormonal activity

A methanol extract of the root (100 g root in methanol 600 ml) did not significantly bind to either estrogen receptor α or β at a concentration of 20.0 µg/ml, but did increase expression of pS2 (an estrogen-sensitive gene) in S-20 cells (*30*).

The expression of the estrogen-regulated gene pS2 in MCF-7 breast cancer cells was assessed after treatment of the cells with a decoction of the roots. The extract and estradiol were equally able to induce RNA expression of pS2, and the extract caused a dose-dependent decrease in cell proliferation ($p < 0.005$) (*31*). The estrogenic effects of a decoction of the roots on the expression of pS2, an estrogen-regulated gene, in breast cancer cell lines MCF-7, T-47D and BT-20 was evaluated by Northern and Western blot analyses. Competitive studies were also performed with ginseng in combination with tamoxifen. The ginseng extract (600.0 µg) and

estradiol (10^{-9} M) induced the expression of pS2 RNA and protein in MCF-7 cells, whereas tamoxifen suppressed expression. Neither the extract nor estradiol induced increased pS2 expression in T-47D or BT-20 cell lines. Although estradiol exhibited a proliferative effect and tamoxifen had an inhibitory effect, the extract of the crude drug had no significant effect on cell proliferation. Because the extract does not exhibit a proliferative effect, it may play a protective role against breast cancer rather than serving as a mitogen (*32*).

Intragastric administration of the powdered root, at a dose of 100.0 mg/kg bw for 28 days, enhanced mounting behaviour of male rats and increased sperm counts in rabbit testes (*33*, *34*). The effect of ginsenoside-Rb1 on the secretion of luteinizing hormone was investigated in vivo and in vitro. Male rats were orchidectomized 2 weeks or subjected to swim training for 1 week before catheterization via the right jugular vein. They were intravenously injected with ginsenoside-Rb1 (10.0 µg/kg bw) or saline 15 minutes before a challenge with gonadotropin-releasing hormone or a 10 minute-swim. Blood samples were collected at several time intervals following intravenous injection of ginsenoside-Rb1. In the in vitro experiment, male rats were decapitated and their anterior pituitary glands were either bisected or enzymatically dispersed. The dispersed anterior pituitary gland cells were primed with dihydrotestosterone for 3 days, and then challenged with ginsenoside-Rbl (10.0 and 100.0 µM, $n = 8$) for 3 h. The concentrations of luteinizing hormone or testosterone in samples were measured by radioimmunoassay. Administration of ginsenoside-Rb1 did not alter the levels of plasma luteinizing hormone in either intact or orchidectomized rats, but significantly increased concentration of plasma luteinizing hormone at the termination of the 10-minute swimming exercise. Administration of ginsenoside-Rb1 resulted in a lower testosterone response to gonadotropin-releasing hormone challenge or swimming exercise than seen in saline-treated rats. Ginsenoside-Rb1 dose-dependently increased the release of luteinizing hormone from both hemi-anterior pituitary gland tissues and the dihydrotestosterone-primed dispersed anterior pituitary gland cells in vitro. These results suggest that ginsenoside-Rb1 increases secretion of luteinizing hormone by acting directly on cells of rat anterior pituitary gland (*34*).

Immune stimulation effects

The immunostimulatory activities of an aqueous extract (1 g in 10 ml water) and a methanol extract (1 g in 200 ml methanol) of the root were assessed in rat alveolar macrophages. Tumour necrosis factor alpha (TNF-α) production was used as a measure of activity. The aqueous extracts (1.0–100.0 µg/ml) significantly stimulated alveolar macrophage TNF-α release. By con-

trast, a methanol extract containing ginsenosides (but no polysaccharides), and pure ginsenoside-Rb1 were not active. Significant TNF-α stimulating activity was found in the extractable polysaccharide fraction (*24*).

The effect of an aqueous extract of the powdered root, containing mainly oligosaccharides and polysaccharides was assessed for immune stimulatory activity in murine spleen cells and peritoneal macrophages. The extract (5.0–500.0 µg/ml) stimulated the proliferation of normal mouse spleen cells, primarily B lymphocytes. The extract activated peritoneal exudate macrophages leading to enhanced production of interleukin-1, interleukin-6, TNF-α and nitric oxide (*35*).

Neurological effects

The neuroprotective effects of a hot aqueous extract of the root (no further description available) on sodium channels were assessed, as neuronal damage during ischaemic episodes has been associated with abnormal sodium fluxes. An aqueous extract of the root was evaluated in tsA201 cells transfected with cDNA expressing alpha subunits of the brain (2a) sodium channel using the whole-cell patch clamp technique. The extract (3.0 mg/ml) tonically and reversibly blocked the sodium channel in a concentration- and voltage-dependent manner. It shifted the voltage-dependence of inactivation by 14 mV (3.0 mg/ml) in the hyperpolarizing direction and delayed recovery from inactivation, whereas activation of the channel was unaffected. Ginsenoside Rb1, a major constituent of the extract, produced similar effects at a concentration of 150 µM (*36*).

Brainstem neurons receiving subdiaphragmatic vagal inputs were recorded in an in vitro neonatal rat brainstem–gastric preparation. Aqueous extracts of the root were added to the gastric compartment or to the brainstem compartment of the bath chamber to evaluate the peripheral gut and central brain effects of the extracts on brainstem unitary activity. After addition of the extract (30.0 µg/ml) to the gastric or brainstem compartment, a concentration-related inhibition of neuronal discharge frequency in the brainstem unitary activity (38.2%, $p < 0.01$) was observed, suggesting that extract may play an important role in regulating the digestive process and modulating brain function (*37*). In a subsequent study, the pharmacological effects of *P. quinquefolius* cultivated in Wisconsin and of *P. quinquefolius* cultivated in Illinois were compared. The results showed that *P. quinquefolius* cultivated in Illinois had both a stronger peripheral gastric and a stronger central brain modulating effect on brainstem neuronal activity. The data indicated that this discrepancy was due to a different ginsenoside profile (*38*).

In a formalin-induced nociception test, mice were force-fed an aqueous extract of the roots for 4 days. Another group of mice were fed with

water as a placebo in a similar fashion. A two-phase formalin test was performed in both groups. Although there was no difference between groups in the first phase, mice treated with the extract spent significantly less time in licking and biting of the injured paws in the second phase, indicating analgesic effects (*39*).

The pseudoginsenoside-F11, an ocotillol-type saponin isolated from the roots has been shown to have antagonistic actions on morphine-induced behavioural changes in mice. Pseudoginsenoside-F11 antagonizes morphine-induced intracellular production of cyclic adenosine monophosphate (cAMP) and antagonizes morphine-induced decreases in dopamine levels in the limbic area of rat. The antagonistic effects of pseudoginsenoside-F11 on methamphetamine-induced behavioural and neurochemical toxicities were studied in mice. Methamphetamine was administered intraperitoneally at a dose of 10.0 mg/kg bw four times at 2-hourly intervals, and pseudoginsenoside-F11 was orally administered at doses of 4.0 and 8.0 mg/kg bw twice at 4-hourly intervals, 60 min prior to methamphetamine administration. The results showed that pseudoginsenoside-F11 ameliorated the anxiety-like behaviour induced by methamphetamine in the light–dark box task, but the change was not statistically significant. In the forced swimming task, pseudoginsenoside-F11 also shortened the prolonged immobility time induced by methamphetamine. In the appetitive-motivated T-maze task, pseudoginsenoside-F11 greatly shortened methamphetamine-induced prolonged latency and decreased the error counts. Similar results were also observed in the Morris water maze task, where administration of pseudoginsenoside-F11 shortened the escape latency prolonged by methamphetamine. There were significant decreases in the contents of dopamine, 3,4-dihydroxyphenacetic acid, homovanillic acid and 5-hydroxyindoacetic acid in the brain of methamphetamine-treated mice (*40*).

Memory effects

The effect of an alcohol extract of the roots on memory was assessed in the scopolamine-induced memory and performance deficits in a spatial learning task. Long-term oral administration of the extract protected against scopolamine-induced amnesia and increased choline uptake in synaptosomal preparations. Treatment with the extract did not alter brain concentrations of norepinephrine, dopamine, 5-HT (serotonin), 3,4-hydroxyphenylacetic acid or 5-hydroxyindoleacetic acid. The extract weakly inhibited the activity of monoamine oxidase in vitro (*41*). Ginsenoside Rb1, a saponin of the crude drug was found to exert beneficial effects on memory and learning, putatively through its actions on the cholinergic system. In situ hybridization studies show that ginsenoside Rb1 increases

the expression of choline acetyltransferase and trkA mRNAs in the basal forebrain and nerve growth factor mRNA in the hippocampus (*42*).

The effect of chronic treatment with ginsenoside Rb1 (5.0 mg/kg/day, intraperitoneal administration for 4 days) was assessed in scopolamine-induced amnesia. The results indicated that treatment with ginsenoside Rb1 can partially attenuate scopolamine-induced amnesia without sedative effects (*43*). In vitro studies show that ginsenoside Rb1 has no effect on quinuclidinyl benzylate binding or on acetylcholinesterase activity, but facilitates the release of acetylcholine from hippocampal slices. The increase in acetylcholine release was associated with an increased uptake of choline into nerve endings; however, calcium influx was unaltered. Thus, the ability of ginsenoside Rb1 to prevent memory deficits may be related to facilitation of acetylcholine metabolism in the central nervous system (*43*). In situ hybridization studies show that ginsenoside Rb1 increases the expression of choline acetyltransferase and trk mRNAs in the basal forebrain and nerve growth factor mRNA in the hippocampus (*42*).

Pharmacokinetics

The pharmacokinetics of ginsenosides A1, A2, B2 and C were studied in rabbits in a one-component open model. Ginsenoside C (protopanaxadiol group ginseng saponin) had a significantly longer half-life, greater plasma protein binding, and lower metabolic and renal clearance than ginsenosides A1, A2 and B2 (protopanaxatriol group ginseng saponins). All ginsenosides except ginsenoside A1 were slowly absorbed after intraperitoneal administration. The ginsenosides were not found in rabbit plasma or urine samples after oral administration. The observed differences in the pharmacokinetics of the ginsenosides may be attributed to differences in protein binding (*44*). Analysis of the ginsenosides and their sapogenins in rabbit plasma and urine was performed. Linear relationships of peak height ratio to weight ratio were obtained for ginsenosides (A1, 20–350 µg; A2, 20–400 µg; B2, 20–300 µg; C, 20–500 µg), and sapogenins (panaxadiol or panaxatriol, 10–200.0 µg) in 0.1 ml (*44*).

Toxicology

The effect of a standardized extract of the roots (4% ginsenosides w/w) and individual ginsenosides (Rb1, Rb2, Rc, Rd, Re, Rf and Rg1) on CYP1 catalytic activities was assessed in vitro. The extract decreased human recombinant CYP1A1, CYP1A2 and CYP1B1 activities in a concentration-dependent manner. The extract was 45-fold more potent than *Panax ginseng* in inhibiting CYP1A2. Rb1, Rb2, Rc, Rd, Re, Rf and Rg1, either separately or as a mixture and at the levels reflecting those found in an inhibitory concentration of 100.0 µg/ml of the extract, did not influence

CYP1 activities. However, at a higher concentration of ginsenoside (50 µg/ml), Rb1, Rb2, Rc, Rd and Rf inhibited these activities. Thus, extracts of the root, which were not treated with calf serum or subjected to acid hydrolysis, inhibited CYP1 catalytic activity, but the effects were not due to Rb1, Rb2, Rc, Rd, Re, Rf or Rg1 (*45*).

Ginsenoside C was more toxic than ginsenoside A2 after intraperitoneal administration to mice. Toxicity was not observed after oral administration of any of the ginsenosides. The genins, panaxadiol and panaxatriol, were more toxic and had larger volumes of distribution than the ginsenosides (*46*).

Clinical pharmacology

Various extracts of the roots, with a specific ginsenoside profile, are reported to decrease postprandial glycaemia. An extract of the roots (containing 1.66% total ginsenosides, 0.9% (20S)-protopanaxadiol ginsenosides and 0.75% (20S)-protopanaxatriol) was assessed on the glycaemic index following a 75 g oral glucose tolerance test in a randomized, single-blind study involving 12 normal volunteers. Each subject received 6 g of the extract orally, or a placebo, 40 minutes before a 75 g oral glucose tolerance test. Venous blood samples were drawn at -40, 0, 15, 30, 45, 60, 90 and 120 min. Repeated measures analysis of variance demonstrated that there was no significant effect of the extract on incremental plasma glucose or insulin or their areas under the curve; indices of insulin sensitivity and release (PI30-0/PG30-0) calculated from the oral glucose tolerance test were also unaffected (*18*).

The effect of escalation of the dose and dosage interval of a root product (500 mg powdered root per capsule) was assessed in individuals who did not have diabetes to determine whether further improvements in glucose tolerance (seen previously when 3 g of crude drug was taken 40 minutes before a 25.0 g glucose challenge) could be attained. Ten healthy volunteers were randomly assigned to receive 0 (placebo), 3, 6 or 9 g of crude drug prepared from the ground root at 40, 80 or 120 minutes before a 25.0 g oral glucose challenge. Capillary blood glucose was measured prior to ingestion of the crude drug or placebo capsules and at 0, 15, 30, 45, 60 and 90 minutes from the start of the challenge. As compared with the placebo, 3, 6 and 9 g of crude drug reduced postprandial incremental glucose at 30, 45 and 60 minutes from the start of the challenge ($p < 0.05$); 3 g and 9 g of the crude drug also did so at 90 minutes. All doses of the crude drug reduced the area under the incremental glucose curve (3 g, 26.6%; 6 g, 29.3%; 9 g, 38.5%) ($p < 0.05$). The crude drug taken at different times did not have any additional influence on postprandial glycaemia (*19*). In a subsequent investigation, 10 patients with type 2 diabetes were randomly

administered 0 g (placebo) or 3, 6 or 9 g of the ground crude drug in capsules at 120, 80, 40 or 0 minutes before a 25 g oral glucose challenge. Capillary blood glucose was measured before ingestion of the crude drug or placebo and at 0, 15, 30, 45, 60, 90 and 120 minutes from the start of the glucose challenge. Two-way analysis of variance (ANOVA) demonstrated that treatment (0, 3, 6 and 9 g crude drug), but not time of administration (120, 80, 40 or 0 min before the challenge) significantly affected postprandial glucose ($p < 0.05$), with significant interaction for area under the curve ($p = 0.037$). Pair-wise comparisons showed that compared with administration of the placebo (0 g), doses of 3, 6 or 9 g of the crude drug significantly ($p < 0.05$) reduced the area under the curve (by 19.7, 15.3 and 15.9%, respectively) and incremental glycaemia at 30 minutes (16.3, 18.4 and 18.4%, respectively), 45 minutes (12.5, 14.3 and 14.3%, respectively), and 120 minutes (59.1, 40.9 and 45.5%, respectively) (*20*).

A preliminary short-term clinical study evaluated the efficacy of the powdered crude drug on postprandial glycaemia in humans. On four separate occasions, 10 subjects who did not have diabetes and nine subjects who had type 2 diabetes mellitus were randomly allocated to receive 3.0 g of the powdered root or placebo capsules, either 40 minutes before or together with a 25.0 g oral glucose challenge. A capillary blood sample was taken during fasting and then at 15, 30, 45, 60, 90 and 120 (only for subjects with type 2 diabetes mellitus) minutes after the glucose challenge. The results of this study demonstrated that in subjects who did not have diabetes, there were no differences in postprandial glycaemia between the subjects who received the placebo and those who received ginseng when administered together with a glucose challenge. When powdered roots were administered 40 minutes prior to the glucose challenge, significant reductions were observed ($p < 0.05$). In subjects with type 2 diabetes mellitus, the same was true whether capsules were taken before or together with the glucose challenge ($p < 0.05$). The reductions in the area under the glycaemic curve were 18% ± 31% for subjects who did not have diabetes and 19 ± 22% and 22 ± 17% for subjects with type 2 diabetes mellitus when capsules were administered before or together with the glucose challenge, respectively (*21*).

A randomized, cross-over study to assess whether a dose of the powdered root of < 3.0 g, administered 40 minutes prior to an oral glucose challenge would reduce postprandial glycaemia in subjects without diabetes was performed. Twelve healthy volunteers received treatment: 0 (placebo), 1, 2 or 3.0 g of the crude drug at 40, 20, 10 or 0 minutes prior to a 25.0 g oral glucose challenge. Capillary blood was collected before administration and at 0, 15, 30, 45, 60 and 90 minutes after the start of the glucose challenge.

Two-way ANOVA showed that the main effects of treatment and administration time were significant ($p < 0.05$). Glycaemia was lower over the last 45 minutes of the test after doses of 1, 2 or 3 g than after placebo ($p < 0.05$); there were no significant differences between the three doses. The reductions in the areas under the curve for these three doses were 14.4 ± 6.5%, 10.6 ± 4.0% and 9.1 ± 6%, respectively. Glycaemia in the last hour of the test and area under the curve were lower when the crude drug was administered 40 minutes before the challenge than when it was administered 20, 10 or 0 minutes before the challenge ($p < 0.05$). Thus, doses of the crude drug within the range of 1–3 g were equally effective (*22*).

Adverse reactions

No adverse reactions have been reported.

Contraindications

Radix Panacis Quinquefolii is contraindicated in cases of known allergy or hypersensitivity to the plant material.

Warnings

No information available.

Precautions

Drug interactions

While no drug interactions have been reported, an extract of the root (containing 10% ginsenosides) inhibited the activity of cytochrome P450 isozymes CYP1A1, CYP1A2 and CYP1B1 in vitro in human liver microsomes (*45*). Thus, there is a potential for interactions with other drugs that are metabolized by these enzymes.

Radix Panacis Quinquefolii and its preparations may lower blood sugar levels. Interactions with antidiabetic drugs are possible, but this subject has not been sufficiently investigated.

Carcinogenesis, mutagenesis, impairment of fertility

At concentrations up to 36.0 mg/ml, neither an aqueous nor a butanol extract of the crude drug was mutagenic in the *Salmonella* microsome assay using *S. typhimurium* strain TM677 in the presence or absence of a metabolic activating mix S9 (*47*).

Pregnancy: non-teratogenic effects

Due to a lack of safety data, use of the crude drug during pregnancy is not recommended.

Nursing mothers
Due to a lack of safety data, use of the crude drug during breastfeeding is not recommended.

Paediatric use
Due to a lack of safety data, use of the crude drug in children under the age of 12 years is not recommended.

Other precautions
No information was found.

Dosage forms
Crude drug, extracts, tinctures and capsules.

Posology
(Unless otherwise indicated)
Oral dose: 3–9 g daily in divided doses (*2*).

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Cortex Phellodendron

Definition
Cortex Phellodendron consists of the dried bark of *Phellodendron amurense* Rupr. or *Phellodendron chinense* Schneid. (Rutaceae) (*1–4*).

Synonyms
No information was found.

Selected vernacular names
Amur cork tree, amur oak, Amurkorkbaum, amurkortrae, amuuri korgipuu, asiatisk kortrae, Chinese cork tree, cork tree, guan huang bo, huang-bo, huangbai, huangbo, hwangbaek, kihada, korkowiec amurski, obaku, oubaku, Siberian cork tree, sikerpe, sikerpe-ni, whang-byeuk-namoo (*5–7*).

Geographical distribution
Found in northern China, Japan, and the Democratic People's Republic of Korea and the Republic of Korea (*6–8*).

Description
A deciduous tree, 10–15 m high, with grey, deeply fissured, corky bark. Leaves opposite, compound-imparipinnate, reaching a length of 30 cm; leaflets 5–13, ovate to ovate-lanceolate, 5–11 cm long by 2.0–3.8 cm wide, tip acuminate, base broadly cuneate rounded, margin obscurely serrate, the underside grayish green, glabrous. Inflorescence terminal panicles, pubescent 5–7 cm across. Flowers, unisexual, dioecious, 4 mm in diameter, yellowish green, sepals, petals and stamens 5–6. Fruit an oval drupe, 1.0 cm in diameter, becoming black when ripe and containing 5 seeds (*6–8*).

Plant material of interest: dried bark
General appearance
Flat or rolled semi-tubular pieces of bark, 2–6 mm thick. Outer surface greyish yellow-brown to greyish brown, even or longitudinally furrowed,

with numerous scars of lenticel. Smooth inner surface yellow to dark yellow-brown, with fine vertical lines. Texture light and hard, fracture fibrous and bright yellow (*1–4*).

Organoleptic properties

Odour: slight; taste: very bitter, mucilaginous and imparts a yellow colour to the saliva on chewing (*1–4*).

Microscopic characteristics

Transverse section reveals a thin yellow outer cortex, scattered with stone cells appearing as yellow-brown dots. The inner cortex is thick, with primary medullary rays expanding its width towards the outer end; the phloem appears as a nearly triangular part between the medullary rays in the secondary cortex, with many medullary rays radiating and gathering at the tip of the triangle. Brown phloem fibre bundles aligned in a tangential direction and crossing over the secondary medullary rays, forming a latticework (*2–4*).

Powdered plant material

Bright yellowish to yellow powder, containing fragments of yellow, thick-walled fibrous bundles or isolated fibres; often accompanied by rows of crystal cells; some groups of stone cells together with idioblasts; fragments of parenchyma cells containing starch grains and oil droplets; fragments of medullary ray and phloem; mucilage cells and mucilage masses; numerous solitary crystals of calcium oxalate, 7–20 μm in diameter; starch grains; simple grains (2–6 μm in diameter) and 2–4 compound grains (*2–4*).

General identity tests

Macroscopic and microscopic examinations, microchemical tests, and thin-layer chromatography (*1–4*).

Purity tests

Microbiological

Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*9*).

Chemical

To be established in accordance with national requirements.

Foreign organic matter

To be established in accordance with national requirements.

Total ash
Not more than 7.5% (*2*, *3*).

Acid-insoluble ash
Not more than 0.5% (*2*).

Water-soluble extractive
Not less than 12% (*10*).

Alcohol -soluble extractive
Not less than 14% (*1*).

Loss on drying
Not more than 9% (*2*, *4*).

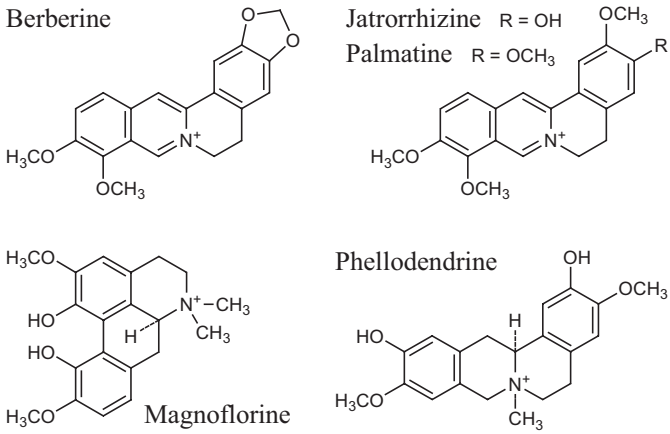
Pesticide residues
The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*11*). For other pesticides, see the *European pharmacopoeia* (*11*) and the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*9*) and pesticide residues (*12*).

Heavy metals
For maximum limits and analysis of heavy metals, consult the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*9*).

Radioactive residues
Where applicable, consult the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*9*).

Chemical assay
Not less than 1.2% berberine by high-performance liquid chromatography (*2*, *3*).

Major chemical constituents
The major constituents are isoquinoline alkaloids, principally berberine (up to 4.75%). Other major alkaloids include magnoflorine (up to 1.1%), palmatine (up to 1.2%), jatrorrhizine (0.5%), as well as phellodendrine and candicine (*5*, *13*, *14*). Structures of berberine, palmatine, jatrorrhizine, magnoflorine and phellodendrine are presented below.



Medicinal uses

Uses supported by clinical data

None.

Uses described in pharmacopoeias and well established documents

Used orally to treat abdominal pain, diarrhoea, gastroenteritis and urinary tract infections (*1*, *15*). While no clinical studies evaluating the safety and efficacy of the crude drug have been reported, clinical studies have been performed to assess the effects of berberine, the major chemical constituent of the crude drug, for the treatment of diarrhoea, ocular trachoma and cutaneous leishmaniasis. Berberine has been shown to be effective for the treatment of bacterially-induced diarrhoea (*16–21*), ocular trachoma and cutaneous leishmaniasis (*22–24*).

Uses described in traditional medicine

Treatment of burns, cough, fever, jaundice, malaria, skin wounds and sores (*5*).

Pharmacology

Experimental pharmacology

An overview of the pharmacological studies evaluating the efficacy of the crude drug and berberine, the major chemical constituent of the plant is presented. The direct applicability of the pharmacological data for berberine to the crude drug is uncertain and needs to be further investigated.

Antidiarrhoeal activity

The infectivity of rotavirus, a virus associated with the development of diarrhoea, was inhibited in vitro by the addition of a decoction of the

crude drug to the culture medium at a concentration of 50.0 µg/ml (*25*). In vitro studies have demonstrated that *Vibrio cholerae* grows in a medium containing berberine, but fails to produce diarrhoea-inducing toxins (*26*). It has been hypothesized that the antidysenteric activity of berberine is due to localized effects on the intestinal tract and not due to bactericidal activity. The suggested mechanism by which berberine exerts its antidiarrhoeal activity is through the activation of α_2-adrenoceptors and by reducing production of cyclic adenosine monophosphate through the inhibition of the activity of adenylate cyclase (*27*), which in turn decreases intestinal motility (*28*). Berberine inhibits in vivo and in vitro intestinal secretions induced by cholera toxin (*29–32*).

Berberine at a concentration of 10.0 mg/loop reduced intestinal secretion stimulated by the heat-labile toxin of *Escherichia coli* in the rabbit ligated intestinal loop model by 70% in situ, and inhibited the secretory response of the heat stable toxin of *E. coli* in mice (*33*, *34*). Berberine at concentrations up to 500.0 µM stimulated ion transport responses in human colonic mucosa that were non-specific for calcium ions or cyclic adenosine monophosphate-mediated signals. In cultured intestinal epithelial monolayers, berberine inhibited calcium and cyclic adenosine monophosphate-mediated responses, indicating that the drug exerts a direct antisecretory effect on the epithelial cells through the inhibition of mucosal chloride secretion (*35*).

Antihypertensive activity

Administration of a lyophilized aqueous extract of the crude drug in drinking-water at a dose of 500.0 mg/kg body weight (bw) to rats for 20 days significantly reduced hypertension induced by heat stress or the administration of metyrapone ($p < 0.01$) (*36*, *37*). Intragastric administration of a 70% ethanol extract of the crude drug at a dose of 600.0 mg/kg bw to rats for 20 days reduced blood pressure (*38*).

Anti-inflammatory activity

A 50% ethanol extract of the crude drug reduced 12-*O*-tetradecanoylphorbol-13-acetate and acetic acid-induced oedema and inhibited oxazolone-induced contact-delayed hypersensitivity ear oedema in mice after application of 0.5 mg/ear (*39*). A 50% methanol extract (1:1) significantly decreased granulation tissues in chick embryo chorioallantoic membranes, indicating anti-inflammatory activity ($p < 0.05$) (*40*). Intragastric administration of berberine to rats with trinitrobenzene sulfonic acid-induced colitis at a dose of 7.5 or 15.0 mg/kg/day reduced histological lesions, morphological damage and myeloperoxidase activity after 1 week of treatment (*41*). Berberine also inhibited the production of the potent in-

flammatory cytokine, interleukin-8, in rectal mucosal cells in vitro at a concentration of 10^{-5} M (*41*). Treatment of a human oesophageal squamous cell carcinoma cell line (YES-2) with berberine (8.0–32.0 µM) for 24 hours reduced the expression of messenger ribonucleic acid for the inflammatory cytokine, interleukin-6 (*42*).

Antimicrobial and antitrypanosomal activities

A hot aqueous extract of the crude drug inhibited the growth of *Escherichia coli* and *Staphylococcus aureus* (*43*); *Streptococcus mutans* (*44*); *Candida albicans*, *C. glabrata* and *C. tropicalis* (*45*). Berberine inhibited the growth of *Helicobacter pylori* in vitro. The median inhibitory concentration ranged from 0.625–40.00 µg/ml (*46*, *47*). Berberine also inhibited the growth of *Staphylococcus aureus* and *Mycobacterium smegmatis*, with a minimum inhibitory concentration of 25.0–50.0 µg/ml (*48*, *49*). Berberine inhibited the growth of *Bacillus subtilis* and *Salmonella enteritidis* in vitro at a concentration of 1.0 mg/ml and 0.5 mg/ml, respectively (*50*). Berberine also inhibited the growth of *Clostridium perfringens* in vitro (150.0 µg/ml), and at a concentration of 1.0 mg/ml significantly inhibited the growth of *Entamoeba histolytica*, *Giardia lamblia* and *Trichomonas vaginalis* and induced morphological changes in the parasites ($p < 0.001$) (*51*). At a concentration of 250.0 µg/ml, a methanol extract of the crude drug inhibited the growth of *Trypanosoma cruzi* in vitro (*52*).

Antiulcer activity

Intragastric administration of a berberine-free fraction of a bark extract (at a dose of 1.0 g/kg bw) reduced ulcer formation in rodents (*15*). The fraction significantly inhibited the formation of ulcers induced by ethanol or aspirin, and of pylorus-ligated ulcer in rats, as well as that of stress ulcer in restrained and water-immersed mice ($p < 0.05$). In addition, secretion of gastric acid in pylorus-ligated rats was significantly reduced by the fraction when administered subcutaneously or intraduodenally, but not when it was administered orally (*15*).

Subcutaneous administration of the same berberine-free fraction to rats at a dose of 20.0 mg/kg bw prevented ethanol-induced ulcers and those induced by pylorus-ligation (*15*). In rats, a hot aqueous extract of the crude drug (dose 100.0 mg/kg bw) inhibited gastric secretion induced by administration of pentagastrin (*53*).

Effects on smooth muscle

Berberine, at a concentration of 1.0 µM, induced relaxation of norepinephrine-precontracted isolated rat aorta (*54*). At a concentration of 10^{-5} M, berberine induced relaxation in isolated precontracted rat mesenteric ar-

teries (*55*, *56*). At a concentration of 0.1 to 100.0 µM, berberine suppressed basal tone and induced a concentration-dependent relaxation of phenylephrine-precontracted rabbit corpus cavernosum in vitro (*57*). Intracavernous injection of berberine to anaesthetized rabbits at a dose of 5.0 mg/kg bw increased intracavernosal pressure from 12.7 to 63.4 mmHg, and the duration of tumescence ranged from 11.5 to 43.7 minutes (*57*).

Immunological effects

The crude drug is reported to have a potent suppressive effect on the cellular immune response. The compounds OB-1 and OB-5 were isolated as the active constituents of a methanol extract of the crude drug, and shown to suppress local graft versus host reactions in mice. OB-1 and OB-5 were identified as the quaternary alkaloids magnoflorine and phellodendrine, respectively. Both compounds suppressed the local graft versus host reaction, when given by intraperitoneal injection to the host mice at a dose of 5.0–20.0 mg/kg bw for 8 consecutive days from the day of spleen cell transfer to cause the reaction. Both OB-1 and OB-5 suppressed delayed-type hypersensitivity induced by picryl chloride, when given by intraperitoneal injection to mice at a dose of 10.0 and 20.0 mg/kg bw for 5 consecutive days from the day of the sensitization, but did not suppress it when given at the time of the challenge (*58*). Phellodendrine suppressed local semisyngeneic graft versus host reactions and systemic allogenic graft versus host reactions in X-ray irradiated mice. Phellodendrine also suppressed the induction phase of delayed-type hypersensitivity induced by sheep red blood cells in mice and tuberculin-induced delayed-type hypersensitivity in guinea-pigs (*59*). Intraperitoneal administration of berberine to mice, at a daily dose of 10.0 mg/kg bw for 3 days before the induction of tubulointerstitial nephritis, significantly reduced pathological injury and improved renal function ($p = 0.001$), as well as decreasing the number of CD3+, CD4+ and CD8+ T-lymphocytes as compared with control animals (*60*).

Toxicology

The oral median lethal dose of berberine in mice was 329.0 mg/kg bw (*61*). An oral dose of berberine (2.75 g) given to dogs produced severe gastrointestinal irritation, profuse watery diarrhoea, salivation, muscular tremors and paralysis. Respiration was not affected. Postmortem analysis of the intestines found them to be contracted, inflamed, and empty or containing mucus and watery fluid. An oral dose of 25.0 mg/kg bw of berberine sulfate induced depression lasting for 6–8 hours; 50.0 mg/kg bw caused salivation and sporadic emesis. A dose of 100.0 mg/kg bw induced persistent emesis and death of all animals 8–10 days later (*62*).

Clinical pharmacology

While no clinical studies evaluating the safety and efficacy of the crude drug could be found, there have been numerous studies evaluating the efficacy of berberine, the major chemical constituent of the plant. Therefore, summaries of these clinical studies have been included in the monograph. The direct applicability of these clinical data to the crude drug is unknown and needs to be further investigated.

In several clinical trials, berberine was effective in the treatment of secretory diarrhoea (*16*–*21*). However, few trials have compared the efficacy of berberine with a positive control, such as tetracycline, in treating fluid loss caused by diarrhoea in patients with cholera or in those with non-cholera diarrhoea (*16*, *17*, *19*, *61*).

A randomized, placebo-controlled, double-blind clinical trial involving 400 patients with acute watery diarrhoea compared the antisecretory and vibriostatic effects of berberine and tetracycline (*16*). Of 185 patients with cholera, those given tetracycline or tetracycline and berberine had a reduced volume and frequency of stools, and duration of diarrhoea. In the group treated with berberine, following oral administration of 100.0 mg four times a day, stool volume was reduced and the concentration of cyclic adenosine monophosphate in the stools was reduced by 77%. Neither berberine nor tetracycline exhibited any benefit over placebo in patients with non-cholera diarrhoea of unspecified etiologies (*16*).

A randomized comparison-controlled trial of 165 patients assessed the antisecretory activity of a 400.0-mg single-bolus dose of berberine sulfate for enterotoxigenic *Escherichia coli*-induced diarrhoea, and either 400.0 mg as a single oral dose or 1200.0 mg of berberine sulfate (400.0 mg every 8 hours) for the treatment of cholera (*19*). In patients with *Escherichia coli*-induced diarrhoea who received a single oral dose of berberine, the mean stool volumes were significantly less than those of controls during three consecutive 8-hour periods after treatment ($p < 0.05$). At 24 hours after treatment, patients with *Escherichia coli*-induced diarrhoea, who were treated with berberine, had a lower stool volume and frequency than patients in the control group (42% versus 20%, $p < 0.05$). Patients with cholera, who received 400.0 mg of berberine, also had a reduction in stool volume, while those treated with 1200.0 mg of berberine plus tetracycline did not. No adverse effects were observed in the patients receiving berberine. The results of this study indicated that berberine was an effective and safe antisecretory drug for treatment of *Escherichia coli*-induced diarrhoea, but had only a modest antisecretory effect in cholera patients, in whom the activity of tetracycline alone was superior (*19*).

Berberine has been used therapeutically for the treatment of cutaneous leishmaniasis, commonly referred to as "oriental sore". It is administered by subcutaneous injection near the site of the lesion (*22–24*). In patients with leishmaniasis caused by *Leishmania tropica*, injection of a preparation containing 2% berberine into the lesions was effective in the treatment of cutaneous leishmaniasis (*22*, *23*).

Adverse reactions

Berberine is well tolerated in humans in therapeutic daily doses of 0.5 g (*61*).

Contraindications

Cortex Phellodendron is contraindicated in cases of hypersensitivity or allergy to the plant material.

Warnings

No information was found.

Precautions

General

No information was found.

Drug interactions

None reported. However, berberine, the major isoquinoline alkaloid occurring in the crude drug, is reported to up-regulate the expression of the human multidrug resistance gene coding for multidrug resistance transporter (pgp-170), thus the treatment of tumours with berberine may result in reduced retention of chemotherapeutic agents such as paclitaxel (*62*, *63*). In a randomized controlled clinical trial, 52 renal transplant recipients were treated with ciclosporin A and 0.2 g berberine three times daily for 3 months, while another 52 subjects received ciclosporin A alone (*64*). For a pharmacokinetic study, six renal transplant recipients were given a 3-mg/kg dosage of ciclosporin A twice daily before and after oral co-administration of 0.2 g berberine three times daily (i.e. berberine was given every 8 hours and cyclosporine every 12 hours) for 12 days. The results of this study showed that the trough blood concentrations and the ratios of concentration:dose of ciclosporin A in the berberine-treated group increased by 88.9% and 98.4%, respectively, compared with those at baseline ($p < 0.05$). Thus, berberine can markedly elevate the blood concentration of ciclosporin A in renal transplant recipients in both clinical and pharmacokinetic studies. This combination may allow a reduction of the dosage of ciclosporin A (*64*).

Furthermore, an ethanol extract of the crude drug inhibited the activity of cytochrome P450 in vitro, with an IC_{50} of < 1% (*65*). Thus, concomitant administration of the crude drug with a drug metabolized via cytochrome P450 may affect drug metabolism (*65*).

Drug and laboratory test interactions
None reported.

Carcinogenesis, mutagenesis, impairment of fertility
A hot aqueous or methanol extract of the crude drug was not mutagenic at a concentration of 100.0 mg/ml in the Ames test in *Salmonella typhimurium* strains TA98 and TA100, or in *Bacillus subtilis* H-17 (Rec+) (*66*).

Berberine was not genotoxic in *Saccharomyces cerevisiae* assessed in the SOS chromotest (*67*). No genotoxic activity was observed with or without metabolic activation, and no cytotoxic or mutagenic effects were seen under non-growth conditions. However, in dividing cells, the alkaloid induced cytotoxic and cytostatic effects in proficient and repair-deficient *Saccharomyces cerevisiae.* In dividing cells, the induction of frameshift and mitochondrial mutations, as well as crossing over, showed that the compound is not a potent mutagen (*67*).

Pregnancy: teratogenic effects
Intragastric administration of a 70% methanol extract of the dried bark to pregnant rats, at a dose of 500.0 mg/kg bw daily from day 13 to the time of delivery did not produce teratogenic effects in the offspring (*68*).

Pregnancy: non-teratogenic effects
Intragastric administration of a 70% methanol extract of the dried bark to pregnant rats, at a dose of 500.0 mg/kg bw daily from day 13 to the time of delivery, did not induce abortion or miscarriage (*68*).

Nursing mothers
Owing to the lack of safety data, the use of Cortex Phellodendron by breastfeeding mothers is not recommended.

Paediatric use
No information was found.

Dosage forms
Crude drug and dried extracts, fluidextracts and tinctures. Store in a tightly sealed container away from heat and light (*3*).

Posology

(Unless otherwise indicated)
Daily dose: 3–10 g daily (*1*).

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Rhizoma Picrorhizae

Definition

Rhizoma Picrorhizae consists of the dried rhizome with root of *Picrorhiza kurrooa* Royle (*1*) or of *Neopicrorhiza scrophulariiflora* Hong [syn. *Picrorhiza scrophulariiflora* Pennell] (Scrophulariaceae) (*2*).

Synonyms

Picrorhiza kurrooa Royle: *Picrorhiza kurroa* Benth., *P. kurroa* Royle ex Benth.

In the formularies and most references, the species is known as *Picrorhiza kurroa* Benth. or *Picrorhiza kurroa* Royle ex Benth. (Scrophulariaceae), but according to the International Code of Botanical Nomenclature article 42, the correct name of the species should be *Picrorhiza kurrooa* Royle (*3*).

Neopicrorhiza scrophulariiflora Hong: *Picrorhiza scrophulariiflora* Pennell.

The rhizomes of *Neopicrorhiza scrophulariiflora* Hong [syn. *Picrorhiza scrophulariiflora* Pennell] are taxonomically similar and have been used in traditional medicine for the same purposes and traded under the same vernacular names (*3*).

Selected vernacular names

Balakadu len, hohwangryun, honglen, hunglen, honkadu, hú huáng lián, kadu, kadugurohini, kalikutki, karu picrorhiza, karru, katki, katu, katuka, katukaa, katuká, katuka rohini, katukarogani, katukarohini, katuki, katuko, katuku rohini, katurohini, katvi, kaur, khanekhaswael, kharbaqe-hindi, kot kaan phraao, koouren, kot kaanphraao, kour, kurri, kuru, kutaki, kutki, kutta, rohini, sutiktaka, tiktarohini, xi zanghu huang lian (*1*, *4–9*).

Geographical distribution

Picrorhiza kurrooa Royle: native to the north-western Himalayas from Kashmir to Sikkim (*1*, *3*, *4*, *6*).

Neopicrorhiza scrophulariiflora Hong: found in the eastern Himalayas to the mountains of Yunan (*3*, *9*).

Description

A perennial herb with a long rhizome. The leaves are basal and alternate, approximately 5–10 cm long. Spikes terminal. Calyx nearly equally in 5 segments. The corolla has 4 or 5 lobes, bilobiate with lobes more or less spreading or nearly actinomorphic. Stamens 4, inserted on corolla tube, slightly didynamous, as long as corolla or strongly exerted. Stigma capitate. Fruit an acute capsule, tapered at top, dehiscing first septicidally and then loculicidally into 4 valves, 12 mm long. Seeds numerous, ellipsoid: seed coat very thick, transparent and alveolate. Pollen grains spheroidal, 3-colpate, with partial or perforate tectum, the partial tectum microreticulate, colpus membrane smooth or sparsely granular (*3*, *10*).

The corolla of *Picrorhiza kurrooa* Royle is 4–5 mm long and 5-lobed, whereas the corolla of *Neopicrorhiza scrophulariiflora* is 9–10 mm long and 4-lobed (*3*).

Plant material of interest: dried rhizome with root

General appearance

Rhizome: 2.5–12.0 cm long and 0.3–1.0 cm thick, subcylindrical, straight or slightly curved, externally greyish-brown, surface rough due to longitudinal wrinkles, circular scars of roots and bud scales and sometimes roots attached, tip ends in a growing bud surrounded by a tufted crown of leaves, in places cork exfoliates exposing dark cortex; fracture, short. Root: thin, cylindrical, 5–10 cm long and 0.5–1.0 mm in diameter, straight or slightly curved with a few longitudinal wrinkles and dotted scars, mostly attached with rhizomes, dusty grey, fracture short, inner surface black with whitish xylem (*1*, *2*, *4*).

Organoleptic properties

Odour: pleasant; taste: bitter (*1*).

Microscopic characteristics

The rhizome portion shows 20–25 layers of cork consisting of tangentially elongated, suberized cells; cork cambium 1–2-layered; cortex single-layered or absent, primary cortex persists in some cases, 1 or 2 small vascular bundles present in the cortex. Vascular bundles surrounded by fibrous bundle sheath. Secondary phloem composed of parenchyma cells and a few scattered fibres. Cambium 2–4-layered. Secondary xylem consists of vessels, tracheids, fibres and parenchyma cells. Vessels vary in size and shape, have transverse oblique articulation; tracheids long, thick-walled, lignified, more or less cylindrical with blunt tapering ends. Starch grains abundant, 25–105 µm in diameter. Root portion, when young,

shows 1-layered epidermis, some epidermal cells elongate forming unicellular hairs. Hypodermis single-layered. Cortex 8–14 layered, consisting of oval to polygonal, thick-walled parenchymatous cells. Primary stele, tetrarch to heptarch, enclosed by a single-layered pericycle and single-layered thick-walled cells of endodermis. Mature roots show 4–15 layers of cork, 1–2 layers of cork cambium. Vessels vary in size and shape, some cylindrical with tail-like, tapering ends; some drum shaped with perforation on end walls or lateral walls. Tracheids cylindrical with tapering pointed ends (*1*).

Powdered plant material

Dusty grey in colour. Groups of cork cell fragments, thick-walled parenchyma, pitted vessels and aseptate fibres. Simple round to oval starch grains, 25–105 µm in diameter (*1*).

General identity tests

Macroscopic and microscopic examinations, microchemical tests and thin-layer chromatography (*1*, *2*), and high-performance liquid chromatography–mass spectrometry (*11*).

Purity tests

Microbiological

Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*12*).

Foreign organic matter

Not more than 2% (*1*).

Total ash

Not more than 7% (*1*, *2*).

Acid-insoluble ash

Not more than 3% (*2*).

Water-soluble extractive

Not less than 20% (*1*).

Alcohol-soluble extractive

Not less than 30% (*2*).

Loss on drying

Contains not more than 13% water (*2*).

Pesticide residues

The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*13*). For other pesticides, see the *European pharmacopoeia* (*13*) and the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*12*) and pesticide residues (*14*).

Heavy metals

For maximum limits and analysis of heavy metals, consult the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*12*).

Radioactive residues

Where applicable, consult the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*12*).

Other purity tests

Chemical tests to be established in accordance with national requirements.

Chemical assays

Total picrosides I and II not less than 9% by high-performance liquid chromatography (*2*).

Major chemical constituents

The major biologically active constituents are iridoid glycosides, cucurbitacin triterpenes and simple phenols. The iridoid glycosides of interest include kutkoside, picrosides I–III, and aucubin, among others. Cucurbitacins B, D, E, F, I and R are among the relevant major triterpenes, and apocynin is a bioactive phenolic constituent of this plant material (*3*, 5, *7*, *15*, *16*). Structures of representative major constituents are presented below.

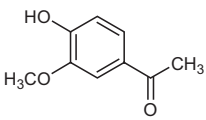
Medicinal uses

Uses supported by clinical data

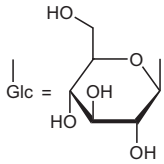
None.

Uses described in pharmacopoeias and well-established documents

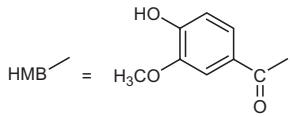
Used orally to treat fever, immune disorders and skin diseases (*1*, *2*). While two studies have suggested a possible role of the rhizome for the treatment of bronchial asthma (*17*, *18*) and viral hepatitis (*19*), no randomized controlled clinical trials have been performed.



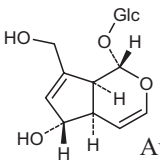
Apocynin



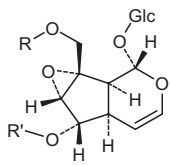
β-D-glucopyranosyl



4-hydroxy-3-methoxybenzoyl

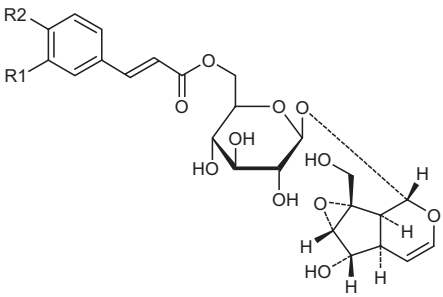


Aucubin



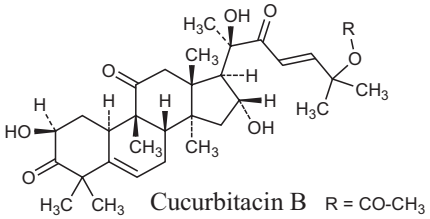
Kutkoside R = HMB, R' = H

Picroside II R = H, R' = HMB



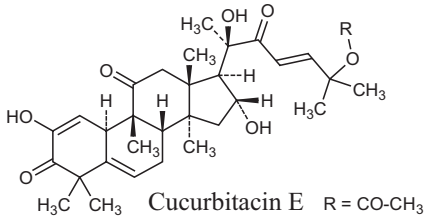
Picroside I R1 = R2 = H

Picroside III R1 = OCH_3, R2 = OH



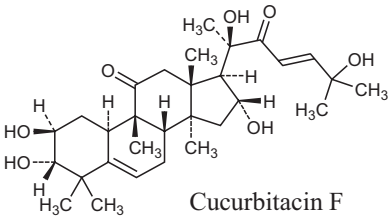
Cucurbitacin B R = CO-CH_3

Cucurbitacin D R = H

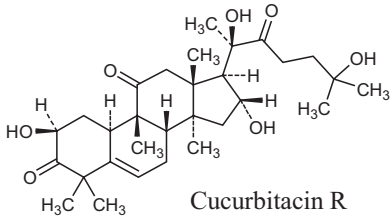


Cucurbitacin E R = CO-CH_3

Cucurbitacin I R = H



Cucurbitacin F



Cucurbitacin R

Uses described in traditional medicine

Used orally to treat anaemia, asthma, diarrhoea, dyspepsia, fever, headaches, obesity, malaria and stomach ache. Also used as an anti-inflammatory agent, a cathartic, a cholagogue and an emmenagogue (*5*).

Pharmacology

Experimental pharmacology

Anti-allergic and anti-asthma activity

Intragastric administration of a dose of 25.0 mg/kg body weight (bw) of a standardized iridoid glycoside fraction from an ethanol extract of the rhizome, inhibited passive cutaneous anaphylaxis in mice (82%) and rats (50–85%) and protected mast cells from degranulation (60–80%) in a concentration-dependent manner. Its effect was also studied in a sensitized guinea-pig ileum preparation in vitro and in normal guinea-pigs in vivo. The Schultz-Dale response was inhibited in sensitized guinea-pig ileum, but histamine-induced bronchospasm was not antagonized or prevented by the fraction, indicating the absence of a direct postsynaptic histamine receptor-blocking activity (*20*).

A dried chloroform (0.1 mg/ml) or dried ethyl acetate extract of the rhizome (0.01 mg/ml) inhibited histamine release in human polymorphonuclear leukocytes treated with rabbit antihuman IgE antibody, or the calcium ionophores A12387 or C5A in vitro (*21*). Intragastric administration of a dried ethyl acetate extract of the crude drug to guinea-pigs at a dose of 10.0 mg/kg bw 1 hour prior to challenge with platelet-activating factor or administration of ovalbumin reduced bronchial obstruction (*21*).

Antidiabetic activity

Intragastric administration of a dried 75% methanol extract of the rhizome (10:1) to rats at a dose of 75.0 mg/kg bw reduced alloxan- or glucose-induced hyperglycaemia (*22*). When administered at a dose of 75.0 mg/kg or 150.0 mg/kg bw, the extract also inhibited the development of alloxan-induced diabetes, and reduced serum peroxide levels and blood urea nitrogen concentration (*22*).

Antihepatotoxic activity

A standardized iridoid glycoside fraction from the rhizome exhibited significant in vitro activity against toxicity induced by thioacetamide (200 µg/ml), galactosamine (400 µg/ml), and carbon tetrachloride (3 µl/ml) in primary cultured rat hepatocytes ($p < 0.01$). Incubation of damaged hepatocytes with the fraction exhibited a concentration-dependent (1–100 µg/ml) curative effect in restoring altered viability parameters (*23*). Intragastric administration of a 95% ethanol extract of the crude drug to rats at a dose of 0.75–6.0 mg/kg bw for 7 days reduced the levels of glutamate oxaloacetate transaminase, glutamate pyruvate transaminase, acid phosphatase, glutamate dehydrogenase and bilirubin in animals with paracetamol- or galactosamine-induced hepatotoxicity (*24*, *25*). The level of glycogen in

the liver was also increased in animals treated with the rhizome extract. Intragastric administration of a methanol extract of the crude drug to mice, at a dose of 670.0 mg/kg bw, suppressed carbon tetrachloride-induced hepatotoxicity (*26*).

Induction of liver injury in 16 mice by injection of carbon tetrachloride (CCl_4) for 9 weeks was reduced by the rhizome. Eight animals were fed the crude drug extract (12.0 mg/kg bw) daily, for 10 days before CCl_4 injection. Control mice (n = 6) were injected with olive oil for the same period. Serum markers of liver injury and histology of liver tissues were studied. Hepatic glutathione, total thiol, glucose-6-phosphate dehydrogenase, catalase, lipid peroxidation and plasma membrane-bound Na^+/K^+ ATPase were also assessed. Feeding the crude drug extract to CCl_4-treated mice reduced serum alanine aminotransferase, aspartate aminotransferase, liver glutathione, catalase and membrane-bound Na^+/K^+ ATPase. Histological lesions of liver and lipid peroxidation were also significantly fewer in these animals (*27*).

Intragastric administration of a dried 75% methanol extract of the powdered rhizomes, at a dose of 150.0 mg/kg bw inhibited the induction of N-nitrosodiethylamine-induced hepatocarcinogenesis and prevented lipid peroxide formation as well as reducing the levels of aniline hydrase and γ-glutamyl transpeptidase (*28*). Intragastric administration of a 95% ethanol extract of the crude drug at a dose of 12.0–25.0 mg/kg bw for 7 days reduced galactosamine and monocrotaline-induced hepatotoxicity (*29*, *30*). The effect of an iridoid glycoside containing an extract of the rhizome on the carcinogenic response and on hepatic and renal antioxidant enzymes of rats treated with 1,2-dimethylhydrazine hydrochloride was investigated (*15*). 1,2-dimethylhydrazine hydrochloride-induced hepatic carcinogenic response and necrosis were inhibited by oral administration of the extract at doses of 40.0 and 200.0 mg/kg bw. Liver γ-glutamyl transpeptidase was reduced to 0.22 ± 0.04 and 0.18 ± 0.03 nmol/mg protein, respectively, by the treatment. Depletion of hepatic and renal antioxidant enzymes such as catalase and superoxide dismutase levels induced by 1,2-dimethylhydrazine hydrochloride was reversed by the treatment, and elevated lipid peroxidation in liver, kidney and serum was reduced. Renal glutathione S-transferase and hepatic glutathione levels which had been depleted by 1,2-dimethylhydrazine hydrochloride were also increased. Oral administration of a standardized iridoid glycoside fraction of the crude drug (25.0 mg/kg bw daily for 15 days), significantly reduced the increases in the activities of tau-glutamyl transpeptidase, 5´-nucleotidase, acid phosphatase and acid ribonuclease, and decreased the activities of succinate dehydrogenase and glucose-6-phosphatase in liver, and the level of

transaminases, sorbitol dehydrogenase, glutamate dehydrogenase, lactate dehydrogenase, acid phosphatase, alkaline phosphatase and bilirubin in the serum of aflatoxin B1-treated rats (*31*).

The mechanism of action of an iridoid glycoside extract of the rhizome on hepatocellular injury and redox status was investigated in a haemorrhage-resuscitation injury in adult rats (*32*). Anaesthetized rats were subjected to haemorrhagic shock by bleeding 30 ml/kg bw. After 60 minutes of shock, rats were resuscitated with twice the shed blood volume of lactated Ringer's solution and were killed 2 hours after resuscitation. Pretreatment with the extract (12 mg/kg bw), given orally for 7 days, resulted in a significant decrease in serum aspartate transaminase and γ-glutamyl transpeptidase levels. The extract also inhibited the lipid peroxidation and nitric oxide release that occurred after haemorrhage-resuscitation and altered the activity of glutathione reductase in a favourable manner. The extract down-regulated the stress-sensitive transcription factor activator protein 1 and decreased the level of c-fos mRNA as well as c-jun and c-fos proteins in liver tissue, indicating that its actions could be mediated through AP1 and associated signal transduction pathways (*32*).

Anti-inflammatory activity

Intragastric administration of a 95% ethanol extract of the rhizome to rats, at a dose of 100.0 mg/kg bw for 3 days, reduced carrageenan-induced pedal oedema (*32*, *33*). Co-administration of the anti-inflammatory antagonists, propranolol or timolol with the 95% ethanol extract of the rhizome reduced its effects.

Choleretic activity

Intragastric administration of a glycoside mixture (picroside-1 and kutkoside 1:1.5, isolated from the rhizome) to guinea-pigs, at a dose of 1.5 mg/kg bw, reduced paracetamol-induced cholestasis by 59.2%; at 3 mg/kg bw, treatment reduced ethynylestradiol-induced cholestasis by 77%. At a dose of 6.0 mg/kg bw the same mixture increased bile flow by 120% (*34*). Intragastric administration of an extract of the crude drug at a dose of 2.0 mg/kg bw increased bile secretion (*35*). Intragastric administration of an unspecified extract (no further details available) of the rhizome to guinea-pigs at a dose of 6.0 mg/kg bw for 7 days prior to thiacetamide administration prevented cholestasis and hepatotoxicity (*37*).

Diuretic activity

Intraperitoneal administration of an aqueous ethanol extract (1:1) of the rhizome to rats at a dose of 250.0 mg/kg bw increased the amount of urine produced over a 4-hour period, in animals preloaded with saline solution (*38*).

Immune stimulatory effects

A dried diethyl ether extract of the rhizome inhibited the classical pathway of complement (median inhibitory concentration (IC_{50}) 1.9 µg/ml). Aqueous, petroleum ether, methanol or ethyl acetate extracts of the crude drug had inhibitory concentrations of 13, 17, 38 and 55 µg/ml, respectively (*3*). The diethyl ether extract of the crude drug also had moderate inhibitory activity against the activation of polymorphonuclear cells (IC_{50} 53 µg/ml) in vitro, and reduced mitogen-induced proliferation of T lymphocytes (IC_{50} 13 µg/ml) (*3*). Two cucurbitacins, picracin and deacetylpicracin, isolated from the rhizomes of the crude drug were responsible for the inhibition of mitogen-induced human T-lymphocyte proliferation (*39*). Incubation of picracin and deacetylpicracin with peripheral blood lymphocytes inhibited interleukin-2 release by phytohaemagglutinin-activated T cells. The IC_{50}s were 5 and 16 µM, respectively. Both picracin and cucurbitacin E also inhibited the release of interleukin 1β and tumour necrosis factor α from monocytes (IC_{50}s were 15 and 3 µM, respectively). Deacetylpicracin was not active at concentrations up to 100 µM (*40*). Intraperitoneal injection of picracin or deacetylpicracin, 1 hour prior to induction of delayed-type hypersensitivity significantly ($p < 0.001$) inhibited the delayed-type hypersensitivity response at doses of 100.0 mg/kg and 30.0 mg/kg bw in mice (*41*).

Toxicology

An aqueous ethanol extract of the rhizome administered intraperitoneally to mice had a median lethal dose of 1.09 g/kg bw, indicating low toxicity (*38*). A 70% methanol extract of the crude drug administered intragastrically to mice exhibited a median lethal dose at > 2 g/kg bw (*41*). An aqueous extract of the rhizome did not stimulate cell proliferation in melanocyte cells in vitro at a concentration of 1 mg/ml (*42*). The median lethal dose of cucurbitacin B was 0.5 mg/kg bw in rabbits after intravenous administration and 340 mg/kg bw of cucurbitacin E in mice after oral administration (*3*). Oral administration of 200 mg/kg bw of an ethanol or diethyl ether extract of the crude drug for 3 or 6 weeks did not increase the weight of the thymus, spleen, liver or mesenterial lymph nodes in mice (*3*).

Clinical pharmacology

In one therapeutic observational study, 20 patients with bronchial asthma were treated with a crude extract of the drug (300 mg three times daily) for 1 year. Outcomes measured included clinical improvement, reduction in the use of bronchodilators and pulmonary function tests. Of the 20 patients treated, 10 showed varying degrees of clinical improvement, including an improvement in pulmonary function tests (*17*).

A preliminary clinical trial involving 36 patients with bronchial asthma assessed the efficacy of an extract of the crude drug. Powdered crude drug at a dose of 75 mg twice daily was administered for 2 weeks. The study reported that 53% of patients had no attacks or chest symptoms, 25% had occasional attacks, which were reduced in severity after treatment. Treatment did not improve the condition of patients who smoked or regularly drank alcohol, and was not effective for acute attacks (*18*).

In a randomized, double-blind placebo-controlled trial involving patients diagnosed with acute viral hepatitis (hepatitis B surface antigen-negative), a powder of the crude drug was administered orally (375 mg three times a day) for 2 weeks (n = 15) or a matching placebo was given (n = 18). The difference in values of bilirubin, serum glutamic oxaloacetic transaminase and serum glutamic pyruvic transaminase between the group that received the placebo and the group that received the treatment with the crude drug was significant. The time in days required for total serum bilirubin to drop to an average value of 2.5 mg% was 75.9 days in patients who received the placebo versus 27.4 days in the treatment group (*19*).

Adverse reactions

No information was found.

Contraindications

No information was found.

Warnings

No information was found.

Precautions

Carcinogenesis, mutagenesis, impairment of fertility

No information was found.

Pregnancy: teratogenic effects

Intragastric administration of a dried 70% methanol extract of the rhizome was not teratogenic in pregnant rats when administered from day 13 of pregnancy onwards at doses of 250.0–500.0 mg/kg bw (*41*).

Pregnancy: non-teratogenic effects

Intragastric administration of a dried 70% methanol extract of the rhizome did not induce abortion in pregnant rats when administered from day 13 of pregnancy onwards at doses of 250–500.0 mg/kg bw (*41*).

Other precautions
No information was found.

Dosage forms
Crude drug, extracts, tablets and tinctures.

Posology
(Unless otherwise indicated)
Oral daily dose: 1–3 g of the powdered crude drug (*1*).

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Oleum Ricini

Definition

Oleum Ricini is the fixed oil obtained by cold expression from the seeds of *Ricinus communis* L. (Euphorbiaceae) (*1–5*).

Synonyms

Ricinus speciosus Burm., *R. viridis* Willd. (*6*).

Vernacular names

Aamudamu, aamudamu chettu, aavanak, African coffee tree, agaliva, amanakku, amidamu, amudam, amudamu, andela, ander, andi, angan-tangan, arand, aranda, arash, aril, arundi, audla, avend, awriwra, ayrunkukri, balambaal olyo, balamball, bedanjir, bele ni vavalagi, bherenda, bimázi, bofareira, botareira, carapate, carrapateira, caster bean plant, caster oil bush, castor, castor-oil plant, castor-oil tree, Cemeiner Wunderbaum, chittmani, 'dan kwasare, diveli, djarak, djarak djeetoon, djarak kaleeki, endaru, endi, eramudapu, erand, eranda, erendi, eri, erund, fampinonoana, franda, gab, gandharva hasthah, harwaa, heran, higuera, higuereta, higuerilla, higuerilla blanca, higuerillo, higuerillo blanco, higuerillo rojo, higuero, himashiyu, himasiyu, ilara, ilarun, jar, kaleeki, kalèkè, karchak, kastalan qajne, kerwa, kesusi, kharwa, kherwa, khirva, khirwa, krapata, kula, kula kula, lahung, lapa-lapa-adete, lara, legezabwende, lepo, lepohina, lepokula, libono, lupono, mahona, masketi, mbono, Mexico weed, mupfure, muriki, noronda, ntoo qaib lab, nyonyo, odagwa, palma Christi, palma de cristo, panchangulam, pei-ma, pomaskwiti, ra' a kau, red chicken tree, red eagle foot, redh, redhi, ricin, ricini oleum virginale, ricino, ricinus, Rizinus, sadabherenda, tangantangan oil plant, tel-enderu, tobsha, tochem-I-bed-anjir, tomontigi, toto ni vavalagi, ttchakkma, txiv taw dlaav laab, udukaju, unapalan, utouto, vatari, verenda, wetapela'celik, wonder tree, wonderboom, zeitel-kharwaa, zurma (*6–16*).

Geographical distribution

Indigenous to tropical regions of Africa, probably native to Ethiopia, but has been adapted and cultivated in subtropical to temperate areas of the world (*6*, *8*, *14*, *17*).

Description

The castor-oil plant is either cultivated or is found growing wild in most tropical and warm temperate countries. It varies from an annual, monoecious herb in temperate regions to a tree attaining a height of 15 m in some tropical areas. There are hundreds of forms of the plant which vary in size, colour of stem and leaves, leaf markings, branching, size, colour and markings of seeds. Shoots and panicles glaucous. Leaves are large, alternate, petiolate, peltate, palmate, 5–12 lobed, the lobes serrate or dentate, green or reddish. The inflorescence is a raceme of staminate and pistillate flowers, often 30–60 cm high, the pistillate flowers occurring above the staminate ones on the floral axis. The fruit is a 3-celled capsule generally covered with soft spines and dehiscing into 3 cocci, each containing an ovoid albuminous seed. Seeds oblong, smooth, mottled and poisonous (*8*, *12*).

Plant material of interest: fixed oil

General appearance

A colourless or pale yellow, clear, viscous oil, miscible with dehydrated ethanol or ether. It is only partly soluble in hexane (a distinction from most other fixed oils). When cooled to 0 °C, it becomes more viscous, and turbidity is gradually formed (*1–5*).

Organoleptic properties

Odour: slight, characteristic; taste: initially bland, with a slightly acrid aftertaste (*3*, *4*).

General identity tests

Physicochemical properties (*2–4*), chemical tests (*3*, *4*), and gas chromatography analysis of fatty acids (*1*).

Purity tests

Microbiological

Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*18*).

Foreign organic matter

To be established according to national requirements.

Total ash

Not applicable.

Acid-insoluble ash
Not applicable.

Water-soluble extractive
Not applicable.

Water
Not more than 0.3% according to the method of Karl-Fischer (*1*).

Pesticide residues
The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*1*). For other pesticides, see the *European pharmacopoeia* (*1*) and the WHO quality control methods for medicinal plant materials (*18*) and pesticide residues (*19*).

Heavy metals
For maximum limits and analysis of heavy metals, consult the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*18*).

Radioactive residues
Where applicable, consult the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*18*).

Other purity tests
Specific gravity d_{25}^{25}: 0.953–0.965 (*1*, *3*, *4*).

Saponification value: 176–187 (*3–5*).

Acid value: not more than 1.5 (*3*, *4*).

Hydroxyl value: 155–177 (*3*, *4*).

Iodine value: 80–90 (*1*, *3*).

Peroxide value: not less than 150 (*1*).

Optical rotation: +3.5 to +6.0 (*1*).

Refractive index: 1.478–1.480 (*5*).

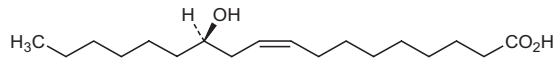
Chemical assays

Fatty acid content: palmitic (2% maximum), stearic (2.5% maximum), oleic and isomers (2.5–6.0%), linoleic (2.5–7.0%), linolenic (2.5% maximum), eicosenoic (1.0% maximum), ricinoleic acid (85–92%) and other fatty acids (maximum) determined by gas chromatography (*1*).

Major chemical constituents

The major chemical constituents of the oil are triacyl glycerols containing an unsaturated and hydroxylated C_{18} fatty acid: (R)-(+)-12-hydroxy-Z-octadec-9-enoic acid, also known as ricinoleic acid (85–92%) (*1*, *17*). It should be noted that ricinoleic acid occurs primarily as its precursor triglyceride, ricinolein (70–77% of the oil) (*20*). Other fatty acids found are: palmitic (2% maximum), stearic (2.5% maximum), oleic and isomers (2.5–6.0%), linoleic (2.5–7.0%), linolenic (2.5% maximum), eicosenoic (1.0% maximum), other acids (1.0% maximum) (*1*). The structure of ricinoleic acid is presented below.

Ricinoleic acid



Medicinal uses

Uses supported by clinical data

None.

Uses described in pharmacopoeias and well established documents

Short-term treatment (3–5 days) for acute constipation when other dietary methods or bulk-forming laxatives have not provided adequate relief. As a cathartic for use in bowel evacuation prior to surgery (*21*). Used externally for topical dermatoses and dermatitis (*5*, *6*).

Uses described in traditional medicine

Used as an emmenagogue, to induce labour, for the treatment of burns, bronchitis, diarrhoea, itching, earache, haemorrhoids, pneumonia, rheumatism and sprains (*6*, *22*, *23*).

Pharmacology

Experimental pharmacology

Anti-inflammatory activity

Pharmacological studies suggest that ricinoleic acid has a similar activity to that of capsaicin, and further suggest a potential interaction of ricinoleic acid with sensory neuropeptide-mediated neurogenic inflammation (*24*).

The pro-inflammatory and anti-inflammatory effects of ricinoleic acid were assessed in an experimental model of blepharitis induced by intradermal injection of carrageenan in guinea-pig eyelids. Topical treatment with ricinoleic acid (10–100.0 mg/guinea-pig) or capsaicin (1–10 mg/guinea-pig) caused eyelid reddening and oedema. At lower doses both drugs

(ricinoleic acid 0.3–3 mg/guinea-pig and capsaicin 0.009–0.09 mg/guinea-pig) significantly potentiated the eyelid oedema induced by carrageenan. The tachykinin NK1 receptor antagonist FK 888 (0.59 mg/kg subcutaneously) reduced carrageenan-induced eyelid oedema induced by either ricinoleic acid or capsaicin. Intravenous administration of the neutral endopeptidase inhibitor, thiorphan (1.3 mg/kg body weight (bw)) significantly enhanced carrageenan-induced eyelid oedema produced by ricinoleic acid. However, repeated topical application of ricinoleic acid (0.9 mg/guinea-pig) or capsaicin (0.09 mg/guinea-pig) for 8 days inhibited carrageenan-induced eyelid oedema. This anti-inflammatory effect was accompanied by a reduction in the tachykinin content of the eyelids, as determined by radioimmunoassay. Ricinoleic acid, like capsaicin, appears to possess dual pro-inflammatory and anti-inflammatory properties; these are observed upon acute and repeated application, respectively (*24*).

A study comparing the anti-inflammatory activities of ricinoleic acid with those of capsaicin in several models of acute and subchronic inflammation was performed. Acute inflammation was induced by intradermal injection of carrageenan in mice or by histamine in the guinea-pig eyelid. Subchronic oedema was induced by injection of complete Freund's adjuvant into the right ventral paw of mice. Tissue substance P was measured in the carrageenan experiments by radioimmunoassay. Acute topical application of ricinoleic acid (0.9 mg/mouse) or capsaicin (0.09 mg/mouse) significantly increased the mouse paw oedema induced by carrageenan, while repeated topical treatment with the same doses of both compounds for 8 days resulted in a marked inhibition of carrageenan-induced paw oedema matched by a reduction in levels of substance P in tissue. Similar effects were found against histamine-induced eyelid oedema in guinea-pigs after acute or repeated application of ricinoleic acid or capsaicin. Ricinoleic acid and capsaicin given for 1–3 weeks reduced the established oedema induced by Freund's adjuvant, a sub-chronic model of inflammation (*25*).

Induction of labour

An investigation exploring the effect of a diet including castor oil on the initiation of labour of pregnant rats was performed. The time of the initiation of labour and course of delivery were observed in rats that received castor oil by gavage on days 18, 19 and 20 of gestation. The diet containing castor oil induced the initiation of labour and shortened the course of the delivery in rats. Ricinoleic acid was the active component of castor oil in this study (*26*).

The effects of a diet containing castor oil on the synthesis of prostaglandin E2 and the mechanism of labour induction were investigated in pregnant rats fed a diet containing castor oil on days 18 and 19 of gesta-

tion. At the time of death (day 20), blood from portal and peripheral veins, and samples from intestinal mucosa, amnion, amniotic cells and placenta were collected, and the tissues were cultured in the presence of ricinoleic acid or indometacin. The concentrations of prostaglandin E2 in the media or blood were measured by radioimmunoassay methods. The prostaglandin E2 levels in the portal vein increased; the prostaglandin E2 levels in peripheral blood showed no significant changes; the prostaglandin E2 levels in the tissues of the intestinal mucosa, placenta, amnion and amniotic cells were increased. Ricinoleic acid stimulated the synthesis of prostaglandin E2 in the tissues in vitro (*27*).

Toxicology

Toxicity studies on castor oil have been performed by incorporating the oil into feed rations at concentrations as high as 10% of the diet. Animals, F344/N rats and B6C3F1 mice, of both sexes were fed the diet for 13 weeks. Exposure to castor oil at dietary concentrations as high as 10% in 13-week studies did not affect survival or body weight gains of rats or mice (10 per sex and dose). No biologically significant effects were noted in haematological analyses in rats. Small increases in total bile acids and in serum alkaline phosphatase were noted at various times during the studies in rats receiving the higher dietary concentrations of castor oil. Liver weights were increased in male rats receiving the 10% dietary concentration and in male and female mice receiving diets containing 5% or 10% castor oil. However, no histopathological lesions were associated with these liver changes, nor were there any compound-related morphological changes in any organ in rats or mice. Thus, no significant adverse effects of castor oil administration were noted (*28*).

Epithelial cells, isolated from small intestine of hamsters, were used to measure in vitro cytotoxicity of the crude drug. The cytotoxicity of ricinoleic acid (castor oil) was assessed by:

- exclusion of trypan blue;
- release of intracellular (prelabelled) 51Cr; and
- inhibition of cellular uptake of 3-*O*-methylglucose.

Ricinoleate produced weak but dose-dependent (0.1–2.0 mM) cytotoxicity as assessed by all three methods (*29*).

In an in vivo study, segments of rabbit ileum were examined by scanning electron microscopy after exposure to various compounds known to stimulate fluid secretion in the small intestine. After perfusion with ricinoleate (castor oil) at a concentration of 10 mM, striking changes were observed at villus tips and on the apicolateral surfaces of villi; erosions of the tips were confirmed by light microscopy of the same pieces of tissue

examined by scanning electron microscopy. The changes that appeared after treatment with ricinoleate were partially reversed during perfusion with control buffer for 2 hours (*30*).

Clinical pharmacology

Laxative effect

Castor oil is an anionic surfactant laxative. Its mechanism of action is primarily as a stool-wetting and stool-softening agent that allows the mixing of water, lipids and other faecal material by altering intestinal permeability and increasing net water and electrolyte secretions. The triglyceride of ricinoleic acid in castor oil is hydrolysed within the small intestine by pancreatic lipases to ricinoleic acid and glycerol. Ricinoleate acts as a local irritant that increases production of cyclic adenosine monophosphate resulting in extensive electrolyte secretion in the small intestine by reducing net absorption of fluid and electrolytes, which ultimately stimulates intestinal peristalsis. Because ricinoleate acts in the small intestine, accumulation of fluid and evacuation takes place rapidly within 1–6 hours, and it continues until the compound is excreted via the colon. Colonic emptying is so complete that several days may elapse before a normal bowel movement occurs (*21*).

To examine the effects of oleic acid and ricinoleic acid on jejunal absorption, steady-state jejunal perfusions were done in healthy volunteers. Taurocholate, used to solubilize the fatty acids, did not influence absorption. Both fatty acids, at a concentration of 10 mM, reversed electrolyte and water net movement and induced reversible fluid secretion. Ricinoleic acid (the active principle of castor oil) was the more potent compound. However, ricinoleic acid was absorbed more slowly than oleic acid, and was associated with higher intraluminal concentrations (*31*).

A retrospective study was performed in patients who underwent elective surgery for colorectal carcinomas. The patients were divided into two groups according to the method of colonic cleansing that was used. Group 1 ($n = 154$) used the traditional bowel preparation which included 30.0 ml castor oil given orally on the day before the operation, in addition to three soap enemas. Patients in group 2 ($n = 36$) were given 500 ml 10% mannitol on the day before the operation. Infections due to wounds developed in 26 patients from group 1 versus 13 patients from group 2. The difference was statistically significant (16.9% versus 36.1%, $p < 0.01$). The differences in the incidence of anastomotic leaks and mortality rate between the two groups were not statistically significant. Castor-oil treatment reduced the number of postoperative infectious wound complications (*32*).

Induction of labour

Castor oil has been widely used as a traditional method of initiating labour in midwifery practice. Its role in the initiation of labour is poorly understood and data examining its clinical efficacy are severely limited. The effects of castor oil administered orally or in enemas for third trimester cervical ripening or induction of labour in comparison with other methods of cervical ripening or induction of labour have been reviewed.

One prospective study of 100 women compared a single dose of castor oil with no treatment. The pregnant women had intact membranes at 40–42 weeks of gestation and were referred for antepartum testing. Inclusion criteria included cervical examination, a Bishop score of 4 or less, and no evidence of regular uterine contractions. Patients were alternately assigned to one of two study groups. One group received a single oral dose of castor oil (60 ml), and the other, no treatment. Castor oil treatment was considered successful if labour began within 24 hours after administration. Groups were compared for onset of labour within 24 hours, method of delivery, presence of meconium-stained amniotic fluid, and Apgar score and birth weight of the infant. Fifty-two women received castor oil and 48 no treatment. Following administration of castor oil, 30 of 52 women (57.7%) began active labour compared to 2 of 48 (4.2%) who had received no treatment. Of the women in whom castor-oil treatment was successful, 83.3% (25/30) of the women delivered vaginally. No difference was found between rates of caesarean section (relative risk (RR) 2.31; 95% confidence intervals (CI) 0.77–6.87). No data were presented on neonatal or maternal mortality or morbidity. There was no difference between either the rate of meconium-stained liquor (RR 0.77; 95% CI 0.25–2.36) or Apgar score < 7 at 5 minutes (RR 0.92; 95% CI 0.02–45.71) between the two groups. The number of participants was small, hence only large differences in outcomes could have been detected. The primary side-effect was nausea. In general, because the trial was small and of poor methodological quality, there is insufficient evidence to support the safety and efficacy of the oil for the induction of labour (*33*, *34*).

Wound-healing effects

Skin graft donor sites are partial-thickness wounds that are commonly managed with gauze-type dressings, which often cause more pain and difficulty in healing than the graft-recipient site. A retrospective study was conducted to ascertain the effects of using a castor oil-balsam of Peru-trypsin-containing ointment on skin graft donor sites in 36 consecutive patients (16 female, 20 male). All donor sites were epithelialized after 11 days (range 6–11 days, mean 8 days) and no wound complications were observed (*35*).

Adverse reactions

Exanthema may occur. In this case, treatment with the oil should be discontinued and should not be used again. Furthermore, castor oil may cause stomach upset, and at higher doses nausea, vomiting, painful abdominal cramps and severe diarrhoea with loss of water and electrolytes. In this case it is necessary to reduce the dosage or to discontinue use of the oil. Continuous use and misuse may increase fluid and electrolyte loss, requiring intravenous rehydration and replacement of electrolytes. Excessive potassium loss may lead to disorders of heart function and muscle weakness (*21*).

A 26-year-old woman, who had had recurrent vulvo-vaginitis for 1 year, was treated with a topical pessary of meclocycline sulfosalicylate. After a 7-day treatment period, the following symptoms appeared: oedema of the labia, widespread irregular abdominal dermatitis in the right iliac fossa and numerous generalized small weals. These symptoms were treated with dexchlorpheniramine maleate without results and it was necessary to prescribe prednisone tablets. Patch tests at 4, 12, 24, 48 and 96 h found a positive response to hydrogenated castor oil, with associated erythema, infiltration and papules after 48 h.

There have been several reports of allergy to castor beans from which castor oil is derived. Cases of asthma have been reported not only in employees in the oil industry, but also in seamen and laboratory workers exposed to the beans. Three important allergens from the crude drug have been identified: a 2S storage albumin of 11 kDa, a 11S crystalloid protein with bands at 50 kDa and a protein doublet of 47 and 51 kDa. These allergens have been named Ric c 1, Ric c 2 and allergen 3. Castor oil extractors, fertilizer workers and farmers may acquire occupational dermatitis from handling plants or beans. Castor oil may be used in cosmetics, particularly lipsticks, because of its superior moisturizing characteristics, and may rarely produce allergic contact dermatitis. Sporadic case-reports of allergy have implicated ricinoleic acid as the causative agent (*36*).

A woman at 39 weeks gestation who had had a previous cesarean delivery had severe abdominal pains and rupture of the membranes shortly after ingesting 5 ml of castor oil. Forty-five minutes later, repetitive variable decelerations prompted a cesarean delivery. During surgery, a portion of the umbilical cord was found to be protruding from a 2-cm rupture of the lower transverse scar (*37*).

Contact dermatitis and a number of cases of chelitis have also been reported following exposure of patients to the crude drug (*38*, *39*).

Contraindications

Oleum Ricini should not be used as a laxative during pregnancy or while breastfeeding (*5*, *13*), or in children under the age of 12 years.

Due to stimulation of bile flow, the oil should not be used by patients with biliary tract obstructions or other biliary disorders.

Oleum Ricini is contraindicated in patients with hypersensitivity or allergy to the oil, appendicitis, chronic inflammatory bowel disorders, undiagnosed abdominal pain or severe dehydration associated with the loss of salt and water.

It should not be used in cases of intestinal blockage and ileus.

Warnings

Induction of labour: use of the crude drug in pregnancy (after 40 weeks of gestation) requires the supervision of a physician, midwife or other experienced health care professional.

As a cathartic: intake of cathartic drugs for more than a few days may lead to a worsening of intestinal hypomotility. Do not take for more than 3–5 days without consulting a health care professional. Overdose may lead to nausea, vomiting, painful abdominal cramps and severe diarrhoea with loss of water and electrolytes. In case of overdose, a health care professional should be consulted regarding possible replacement of fluid and electrolytes.

Precautions

Drug interactions

Concomitant use of castor oil with cardiac glycosides, antiarrhythmic drugs, diuretics, cortisol, liquorice, antihistamines and fat-soluble vitamins may reduce the efficacy of these drugs or, in the case of cardiac glycosides, may increase the risk of adverse events due to fluid and electrolyte losses.

Carcinogenesis, mutagenesis, impairment of fertility

Genetic toxicity studies were negative for induction of mutations in the Ames test using *Salmonella typhimurium* for induction of sister chromatid exchanges or chromosomal aberrations in Chinese hamster ovary cells, and for induction of micronuclei in the peripheral blood erythrocytes of mice fed a diet containing 10% seed oil for 13 weeks (*28*). No significant changes were noted in a screening study for male reproductive end-points, including sperm count and motility, and no changes were observed in the

length of estrous cycles of rats or mice given diets containing castor oil at a concentration of up to 10% (*28*).

Pregnancy: non-teratogenic effects
Oleum Ricini as a laxative is contraindicated in pregnancy before 40 weeks of gestation, due to potential induction of labour.

Nursing mothers
Oleum Ricini is contraindicated in breastfeeding mothers. Ricinoleic acid is absorbed into the bloodstream and is excreted into human breast milk. In suckling infants it has a purgative effect (*13*).

Paediatric use
See Contraindications.

Other precautions
No information was found.

Dosage forms
Fixed oil, capsules (*8*).

Storage
Preserve in well-closed containers, protected from light.

Posology
(Unless otherwise indicated)
As a laxative: single daily dose of 1–10 ml (*40*). Dose for induction of labour: under medical supervision, maximum single dose of 4–60 ml (*33*, *34*). Topical use: fixed oil (*5*).

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Aetheroleum Rosmarini

Definition
Aetheroleum Rosmarini consists of the essential oil, obtained by steam distillation, from the flowering aerial parts of *Rosmarinus officinalis* L. (Lamiaceae) (*1*).

Synonyms
No information was found.

Selected vernacular names
Alecrim, azir, biberine, biberye, boithran, common rosemary, echter Rosmarin, encensier, garden rosemary, gusmarino, hasalban, hatsa louban, hhassâ lubân, iklil, iklil el jabal, iklil kuhi, kusdilli, mannenrou, old man, romani, romarin, romero, romero blanco, rosmariin, rosmarina, Rosmarin, rosmarini, rosmarino, rosemary, tresmarino (*2–4*).

Geographical distribution
Native to Mediterranean region of Europe, and cultivated worldwide (*4–7*).

Description
A bushy, low, much branched, perennial sub-shrub attaining a height of about 1 m. Leaves leathery with fringed margin, 1.0–2.5 cm long, aromatic, evergreen, opposite, sessile, linear and coriaceous. Old branches brown in colour. Spiciform inflorescences of pale blue or light lilac flowers spotted with purple, with the two stamens projecting far beyond the corolla (*4*, *5*, *7*).

Plant material of interest: essential oil
General appearance
Clear, mobile, colourless to pale yellow liquid (*1*).

Organoleptic properties
Odour: characteristic (*1*).

Microscopic characteristics
Not applicable.

Powdered plant material
Not applicable.

General identity tests
Physicochemical properties, thin-layer and gas chromatography (*1*).

Purity tests
Microbiological
Not applicable.

Foreign organic matter
To be established in accordance with national requirements.

Total ash
Not applicable.

Acid-insoluble ash
Not applicable.

Water-soluble extractive
Not applicable.

Loss on drying
Not applicable.

Pesticide residues
The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*1*). For other pesticides, see the *European pharmacopoeia* (*1*) and the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*8*) and pesticide residues (*9*).

Heavy metals
For maximum limits and analysis of heavy metals, consult the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*8*).

Radioactive residues
Where applicable, consult the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*8*).

Other purity tests

Relative density: 0.895–0.920 (*1*).
Refractive index: 1.464–1.473 (*1*).
Optical rotation: -5° to + 8° (*1*).
Acid value: not more than 1.0 (*1*).

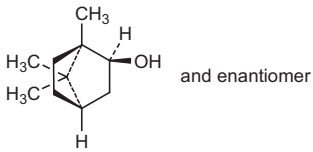
Chemical assays

Gas chromatographic analysis of Spanish type rosemary oil: α-pinene (18–26%), camphene (8–12%), β-pinene (2.0–6.0%), β-myrcene (1.5–5.0%), limonene (2.5–5.0%), 1,8-cineol (16.0–25.0%), *p*-cymene (1.0–2.2%), camphor (13.0–21.0%), bornyl acetate (0.5–2.5%), α-terpineol (1.0–3.5%), borneol (2.0–4.5%) and verbenone (0.7–2.5%) (*1*).

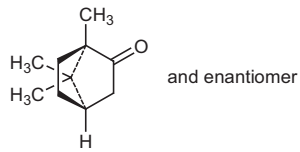
The oil of rosemary from Morocco and Tunisia contains: α-pinene (9–14%), camphene (2.5–6.0%), β-pinene (4.0–9.0%), β-myrcene (1.0–2.0%), limonene (1.5–4.0%), 1,8-cineol (38.0–55.0%), *p*-cymene (0.8–2.5%), camphor (5.0–15.0%), bornyl acetate (0.1–1.5%), α-terpineol (1.0–2.6%), borneol (1.5–5.0%) and verbenone (0.4%) (*1*).

Major chemical constituents

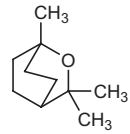
The chief constituents of rosemary oil are camphor (5–31%), 1,8-cineol (15–55%), α-pinene (9–26%), borneol (1.5–5.0%), camphene (2.5–12.0%), β-pinene (2.0–9.0%), limonene (1.5–5.0%), verbenone (2.2–11.1%), β-caryophyllene (1.8–5.1%) and myrcene (0.9–4.5%) (*1*, *10*, *11*). The structures of 1,8-cineole, borneol and camphor are presented below.



Borneol



Camphor



1,8-Cineole

Medicinal uses

Uses supported by clinical data

None.

Uses described in pharmacopoeias and well established documents

Used orally for the treatment of dyspeptic complaints, and in external applications for supportive management of rheumatic complaints and circulatory disorders (*12*, *13*). Although one pilot study has indicated that the

crude drug may enhance cognition (*13*), further data from randomized controlled clinical trials are required before any therapeutic recommendations can be made.

Uses described in traditional medicine

Used as a cholagogue, diaphoretic, digestant, diuretic, emmenagogue, laxative and a tonic (*3*, *5*, *6*). Also used in the management of headache, menstrual disorders, nervous menstrual complaints, tiredness, defective memory, sprains and bruises (*14*).

Pharmacology

Experimental pharmacology

Antihepatotoxic activity

The hepatoprotective and antimutagenic effects of the essential oil were compared to those of ethanol leaf extracts in vivo using carbon tetrachloride (CCl_4) and cyclophosphamide as the hepatotoxic and mutagenic compounds. The results of the study showed that intragastric administration of 1.5 g/kg body weight (bw) of the essential oil or the leaf extract to rats for 3 weeks protected the animals against CCl_4-induced hepatotoxicity. The results were similar to those obtained using the control compound, silymarin (*15*). The results showed amelioration of most of the serum and liver parameters studied, and were confirmed by histopathological examination of the liver tissue. The hepatoprotectant activity of essential oil was not as great as that of the leaf extract. However, intragastric pretreatment of mice for 7 days with the essential oil (1.1 mg/g bw), reduced cyclophosphamide-induced mitodepression in bone marrow cells (*15*).

Antimicrobial activity

The essential oil weakly inhibited the growth of *Acinetobacter iwoffi*, *Bacillus subtilis*, *Erwinia carotovora*, *Shigella flexneri*, *Staphylococcus aureus*, *Streptococcus pyogenes and Yersinia enterocolitica* when added undiluted or in a 1:1 dilution to the agar medium (*16*).

Antispasmodic effects

The essential oil inhibited electrically stimulated muscular contractions in isolated guinea-pig ileum, but the effect of the lowest spasmolytic dose is preceded by an initial increase in the electrically stimulated contractile response. Pinene, which had weak spasmogenic properties, was considered to be the active constituent (*17*). In guinea-pig ileum, addition of the essential oil to the bath medium inhibited acetyl choline-induced contrac-

tions, with a median inhibitory concentration (IC_{50}) of 465 nl/ml of the oil and 41 nl/ml of 1,8-cineole (*18*).

Osteoclastic effects

The essential oil, and its individual constituents, camphor, borneol, thymol, α-pinene, β-pinene and bornyl acetate, inhibited bone resorption when added to the food of rats. The monoterpenes borneol, thymol and camphor, were directly inhibitory in the osteoclast resorption pit assay. Within 30 minutes, borneol inhibited the formation of actin rings, a characteristic of resorbing osteoclasts indicating cell polarization. Both the in vitro and the in vivo effects of borneol were reversible (*19*).

Enzyme induction

Modulation of cytochrome P450 isozymes and detoxification enzymes was compared after oral administration of the dried leaves, a dried dichloromethane leaf extract or the essential oil to rats. The animals received the leaves, extracts or essential oil in their rations at a concentration of 0.5% (w/w) for 2 weeks. The effects of the treatments were evaluated in assays for cytochrome P450 isozymes 1A, 2B, 2E1, glutathione S-transferase, nicotinamide adenine dinucleotide phosphate (NAD(P)H), quinone reductase and uridine diphosphate (UDP)-glucuronosyltransferase activities and on protein levels (immunoblot analyses). The results demonstrated that the essential oil selectively induced cytochrome P450, particularly isozyme 2B. The leaf extract enhanced both cytochrome P450 and detoxification enzymes. A dichloromethane extract of the leaves acted as a monofunctional inducer, inducing glutathione S-transferase, quinone reductase and UDP-glucuronosyltransferase, in particular UDP-glucuronosyltransferase 1A6 (*20*).

Toxicology

The embryotoxic effects of d-camphor were investigated in rats and rabbits after intragastric administration for the treatment of hypotonic circulatory dysregulations. No evidence of teratogenicity was observed when d-camphor was administered orally to pregnant rats during the fetal period of organogenesis, at doses up to 1 g/kg bw per day, and to pregnant rabbits at doses up to 681 mg/kg bw per day. For rats, the no-observed-effect level for the fetal organism was above 1 mg/kg bw, and for rabbits, above 681 mg/kg bw. In rat dams a dose-dependent reduction in food intake and salivation was noted at doses of 464 mg/kg bw and higher. The high dose of 1 mg/kg bw per day resulted in pronounced signs of toxicity such as clonic convulsion, pilo-erection, reduced motility and body weight gain. In rabbit dams, intragastric administration of a high dose (681 mg/kg

bw per day) resulted in reduced body weight gain and food consumption. No increased incidence of variations, retardations or malformations was observed in any of the offspring at any of the doses, not even at the highest dose tested (rat: 1000 mg/kg bw per day; rabbit: 681 mg/kg bw per day). The daily maximum dose of camphor for human therapeutic use is approximately 1.43 mg/kg bw. Hence, under the present test conditions the therapeutic ratio is above 450 for the end-point of embryotoxicity, reflecting a wide margin of safety (*21*). Since d-camphor is only one of the chemical constituents of the essential oil, the relevance of these data to the toxicology of the essential oil needs to be investigated.

Clinical pharmacology

A clinical study to assess the olfactory impact of the essential oils of lavender (*Lavandula angustifolia*) and rosemary (*Rosmarinus officinalis*) on cognitive performance and mood in healthy volunteers was performed (*13*). One hundred and forty-four participants were randomly assigned to one of three independent groups. Each of the subjects was asked to take the Cognitive Drug Research (CDR) computerized cognitive assessment battery in a cubicle containing one of the two odours or no odour (control). Visual analogue mood questionnaires were completed before exposure to the odour and after completion of the test battery. The participants were misled as to the genuine aim of the study until the completion of testing to prevent expectancy effects from possibly influencing the data. The outcome variables from the nine tasks that constitute the CDR core battery feed into six factors that represent different aspects of cognitive functioning. Analysis of performance revealed that lavender produced a significant decrement in performance of working memory, and impaired reaction times for both memory and attention-based tasks compared to the performance of the controls. In contrast, rosemary produced a significant enhancement of performance for overall quality of memory and secondary memory factors, but also impaired speed of memory compared to controls. With regard to mood, comparisons of the change in ratings from baseline to post-test revealed that following the completion of the cognitive assessment battery, both the control group and the group exposed to lavender were significantly less alert than the subjects exposed to rosemary; however, subjects in the control group were significantly less content than subjects who received the rosemary and lavender treatments (*13*).

Frontal electroencephalogram asymmetry shifting from baseline was examined in adults and infants exposed to lavender and rosemary oils by re-analysing previously published data from two studies. The results from 39 adults revealed significant electroencephalogram shifting in the lavender-treated group, with greater relative left frontal electroencephalogram

activation (associated with greater approach behaviour and less depressed affect). The participants in the two groups exposed to the two aromas were further grouped by those with greater baseline, relative to left frontal electroencephalogram activation versus those with a greater baseline relative to right frontal activation. Collapsing across aroma groups, those with greater baseline, relative to right frontal activation, shifted left during exposure to the aroma. Those with greater baseline relative to left frontal activation did not change. In the group subjected to the rosemary aroma, those with greater baseline relative to right frontal electroencephalogram activation shifted left during exposure to the aroma, while those with greater baselines relative to left frontal electroencephalogram activation shifted right. The second study, involving 27 full-term newborns revealed no significant shifts in asymmetry in response to either aroma. However, when the aroma groups were collapsed, the right frontal electroencephalogram group exhibited significant shifting relative to left frontal electroencephalogram activation. This finding was similar to the findings in adults, suggesting that both lavender and rosemary may induce left frontal electroencephalogram shifting in adults and infants who show greater baselines relative to right frontal electroencephalogram activation (*22*).

The effects of lavender and rosemary oils on electroencephalogram activity, alertness and mood were assessed in 40 adults given 3 minutes of aromatherapy. Participants were also given simple mathematical computations to do before and after the therapy. The lavender-treated group showed increased beta power, suggesting increased drowsiness; they also had less depressed mood and reported feeling more relaxed and performed the mathematical computations faster and more accurately following aromatherapy. The rosemary-treated group, on the other hand, showed decreased frontal alpha and beta power, suggesting increased alertness. They also had lower anxiety scores, reported feeling more relaxed and alert and they were only faster, not more accurate, at completing the mathematical computations after the aromatherapy session (*23*).

Adverse reactions

Following oral use gastrointestinal complaints and hypersensitivity reactions may occur rarely. Inhalation can occasionally cause irritation and very rarely laryngospasm (*24*).

External use may worsen bronchospasm. Rarely hypersensitivity reactions of the skin may occur. Photoaggravated allergic contact dermatitis (*25*, *26*) and cheilitis (*27*) have been reported.

Contraindications

Aetheroleum Rosmarini is contraindicated in cases of hypersensitivity or allergy to the plant material. It should not be used in patients suffering from bronchial asthma or bronchitis or on damaged skin, such as in cases of burns, lesions or skin rashes.

Warnings

Due to its irritant properties, the essential oil should not be used on the face or mucosa, and contact with the eyes should be avoided. After application of the essential oil, wash hands to avoid accidental contact with the face and eyes. As with all essential oils, do not exceed the recommended dose.

If there is persistence or worsening of rheumatic symptoms, e.g. in cases of redness, swelling or over-heating of joints, patients should seek advice from a health care practitioner.

Precautions

Drug interactions

Although drug interactions have not been reported, cineole, the main constituent of the oil is known to induce liver metabolic enzymes in animals. Therefore, the oil may interact with other prescription medications.

Carcinogenesis, mutagenesis, impairment of fertility

The crude drug is anti-mutagenic in rats treated with cyclophosphamide (*15*).

Pregnancy: teratogenic effects

See Toxicology.

Pregnancy: non-teratogenic effects

See Toxicology.

Nursing mothers

Due to the lack of safety data, the use of the crude drug during breastfeeding is not recommended.

Paediatric use

Due to the lack of safety data, administration of the crude drug to children under the age of 12 years is not recommended.

Other precautions

No information was found.

Dosage forms

Essential oil for oral and external use (*24*) and aromatherapy (*13*, *22*). Store in a cool place in an airtight container, protected from light (*28*).

Posology

(Unless otherwise indicated)
Daily dosage for oral administration: 1 ml of essential oil (*24*).
External use: 6–10% essential oil in semi-solid and liquid preparations (*24*).

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Folium Rosmarini

Definition
Folium Rosmarini consists of the whole dried leaves of *Rosmarinus officinalis* L. (Lamiaceae) (*1*).

Synonyms
No information was found.

Selected vernacular names
Alecrim, azir, biberine, biberye, boithran, common rosemary, echter Rosmarin, encensier, garden rosemary, gusmarino, hasalban, hatsa louban, hhassâ lubân, iklil, iklil el jabal, iklil kuhi, kusdilli, mannenrou, old man, romani, romarin, romero, romero blanco, rosmariin, Rosmarin, rosmarina, rosmarini, rosmarino, rosemary, tresmarino (*2–4*).

Geographical distribution
Native to the Mediterranean region of Europe, and cultivated worldwide (*4–7*).

Description
A bushy, low, much branched, perennial sub-shrub attaining a height of about 1 m. Leaves leathery with fringed margin, 1.0–2.5 cm long, aromatic, evergreen, opposite, sessile, linear and coriaceous. Old branches brown in colour. Spiciform inflorescences of pale blue or light lilac flowers spotted with purple, with the two stamens projecting far beyond the corolla (*4*, *5*, *7*, *8*).

Plant material of interest: dried leaves
General appearance
Leaves linear to linear-lanceolate, curved, 1–4 cm long, 2–4 mm wide; coriaceous, greyish-green or occasionally brownish; margins entire and strongly revolute, apex obtuse, base tapering and non-petiolate; upper

surface dark green, reticulately pitted, lower surface tomentose. Occasional pieces of stems up to 4 cm long, 1–2 mm wide, dark brown to greenish, tomentose or woody with numerous opposite and decussate leaf scars (*1*).

Organoleptic properties

Odour: strongly aromatic; taste: pungently aromatic, camphoraceous and bitter (*1*).

Microscopic characteristics

Leaf dorsiventral; upper epidermal cells polygonal with slightly thickened walls and occasional pits; lower epidermal cells sinuous; numerous diacytic stomata on the lower surface only; very abundant uniseriate, multicellular, much-branched covering trichomes on the lower epidermis, also glandular trichomes with a unicellular stalk and unicellular, bicellular or multicellular head occurring on both epidermises; hypodermis underlying the upper epidermis composed of large, ovoid cells with thickened and beaded anticlinal walls; these cells extending across the lamina at intervals, separating the two-layered palisade into large, crescent-shaped areas, each with a group of spongy mesophyll (*9*).

Powdered plant material

Greyish-green to yellowish-green. Shows fragments of lower epidermis with straight to sinuous-walled cells and numerous diacytic stomata; fragments of the upper epidermis with straight-walled cells, slightly thickened and pitted, and an underlying hypodermis composed of large, irregular cells with thickened and beaded anticlinal walls; fragments in sectional view showing the hypodermal cells extending across the lamina at intervals, separating the one or two-layered palisade into large, crescent-shaped areas; numerous multicellular, extensively branched, covering trichomes of the lower epidermis and rare conical covering trichomes of the upper epidermis; glandular trichomes of 2 types, the majority with a short, unicellular stalk and a radiate head composed of 8 cells, others, less abundant, with a unicellular stalk and a spherical, unicellular or bicellular head. Occasional cork fragments, fibres, vascular tissue and lignified parenchyma from the stems (*1*).

General identity tests

Macroscopic and microscopic examinations (*1*), thin-layer chromatography (*1*), and high-performance liquid chromatography for phenolic acids (*10*).

Purity tests

Microbiological

Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*11*).

Foreign organic matter

Not more than 5% of stem, and not more than 2.0% other foreign matter (*1*).

Total ash

Not more than 9.0% (*1*).

Acid-insoluble ash

Not more than 1.5% (*9*).

Water-soluble extractive

Not less than 15.0% (*9*).

Water content

Not more than 10% (*1*).

Pesticide residues

The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*1*). For other pesticides, see the *European pharmacopoeia* (*1*) and the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*11*) and pesticide residues (*12*).

Heavy metals

For maximum limits and analysis of heavy metals, consult the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*11*).

Radioactive residues

Where applicable, consult the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*11*).

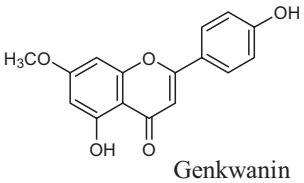
Chemical assays

Not less than 1.2% (v/w) of essential oil and not less than 3% total hydroxycinnamic acid derivatives expressed as rosmarinic acid (*1*).

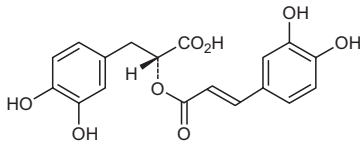
Major chemical constituents

Contains up to 2.5% of essential oil, the chief constituents of which are camphor (5–21%), 1,8-cineole (15–55%), α-pinene (9–26%), borneol (1.5–5.0%),

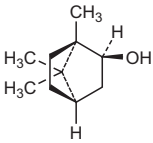
camphene (2.5–12.0%), β-pinene (2.0–9.0%) and limonene (1.5–5.0%). Phenolic compounds are represented by flavonoids with a methylated aglycone (e.g. genkwanin) and by phenolic acids (> 3%), particularly by rosmarinic, chlorogenic and caffeic acids. Also present are tricyclic diterpenes such as rosmaridiphenol, carnosol, carnosic acid and rosmanol, and diterpenes, including seco-hinokiol (*1*, 3, 5, *7*, *13*, *14*). The structures of rosmarinic acid, 1,8-cineole, borneol, camphor, genkwanin and carnosol are presented below.



Genkwanin

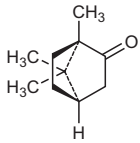


Rosmarinic acid



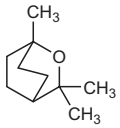
and enantiomer

Borneol

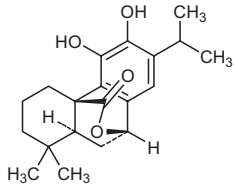


and enantiomer

Camphor



1,8-Cineole



Carnosol

Medicinal uses

Uses supported by clinical data

None.

Uses described in pharmacopoeias and well established documents

Orally as a carminative and spasmolytic to treat dyspepsia (*9*). Externally for supportive therapy of rheumatism and circulatory disorders (*13*).

Uses described in traditional medicine

Orally as a cholagogue, diaphoretic, diuretic, emmenagogue and as a tonic (*3*, *6*, *7*). Also used in the management of headache, menstrual disorders, nervous menstrual complaints, tiredness and defective memory. Used externally for treatment of spraining and bruising (*15*).

Pharmacology

Experimental pharmacology

Antihepatotoxic activity

Intragastric administration of 200.0 mg/kg body weight (bw) of a standardized methanol extract of the leaves (corresponding to 6.04 mg/kg bw

of carnosol) to rats, 1 hour after treatment of the animals with carbon tetrachloride (CCl_4), fully prevented CCl_4-induced lipid peroxidation in the liver. The CCl_4-induced increase in plasma bilirubin concentrations and alanine aminotransferase activity was completely normalized after treatment with the extract. The treatment also resulted in a significant recovery from CCl_4-induced decrease in liver glycogen content. The extract also increased liver cytosolic reduced glutathione activity and produced an additional increase in plasma glutathione activity in rats treated with CCl_4. Histological evaluation showed that the extract partially prevented CCl_4-induced inflammation, necrosis and vacuolation (*16*).

The hepatoprotective and antimutagenic effects of an ethanol extract of the leaves were investigated using CCl_4 and cyclophosphamide as the hepatotoxic and mutagenic compounds. The results indicated that intragastric administration of the ethanol extract (1.5 g/kg bw) to rats for 3 weeks produced a pronounced hepatoprotective effect as compared with silymarin (reference compound) due to the amelioration of most of the serum and liver parameters studied and confirmed by histopathological examination of the liver tissue. Pretreatment of mice for 7 days with the essential oil (1.1 mg/g bw) followed by intraperitoneal administration of cyclophosphamide significantly reduced the induced mitodepression in the bone marrow cells of the animals (*17*).

Anti-inflammatory activity

Treatment of mouse macrophage RAW 264.7 cells with carnosol markedly reduced lipopolysaccharide-stimulated nitric oxide production in a concentration-related manner with an IC_{50} of 9.4 µM. Western blot, reverse transcription–polymerase chain reaction, and northern blot analyses demonstrated that carnosol decreased lipopolysaccharide-induced inducible nitric oxide synthase mRNA and protein expression. Carnosol treatment reduced the translocation of nuclear factor-kappa B subunits and the binding activity of nuclear factor-B DNA in activated macrophages. Carnosol also inhibited inducible nitric oxide synthase and nuclear factor-B promoter activity in a transient transfection assay. These activities were due to down-regulation of inhibitor B kinase activity by carnosol (5 µM), which in turn inhibited lipopolysaccharide-induced phosphorylation as well as degradation of inhibitor B. Carnosol also inhibited lipopolysaccharide-induced p38 and p44/42 mitogen-activated protein kinase activation at a higher concentration (20 µM). These results suggest that carnosol suppresses production of nitric oxide and inducible nitric oxide synthase gene expression by inhibiting activation of nuclear factor-B, and provide possible mechanisms for its anti-inflammatory activity (*18*).

Antioxidant activity

Oxidative damage to DNA, RNA, proteins and cell membranes occurs when the cellular concentration of reactive oxygen species exceeds the capacity of the cell to eliminate them. The crude drug and its constituents have been shown to have both antioxidant and pro-oxidant properties both in vitro and in vivo (*19–21*). The antioxidant activity of six extracts of the leaves, with different compositions of six polyphenolic compounds (carnosic acid, carnosol, 12-*O*-methylcarnosic acid, rosmarinic acid, isoscutellarein 7-*O*-glucoside and genkwanin), were tested in aqueous (malonyldialdehyde formation) and lipid systems (Rancimat method). The results demonstrate that the extracts were excellent antioxidants in both systems at a concentration of 500 ppm (*22*). The ability of the phenolic diterpenes, carnosol, rosmanol and epirosmanol, to prevent oxidization of low-density lipoprotein in human blood and their ability to scavenge free radicals and superoxide anions was assessed in vitro. The antioxidant activities were evaluated by the thiobarbituric acid reactive substances (TBARS) assay and electron spin resonance method. The inhibition of the $Cu2^{+}$-mediated oxidization of apo B formation in low-density lipoprotein was investigated by fluorescence spectroscopy. The results demonstrated that carnosol, rosmanol and epirosmanol inhibited lipid peroxidation and oxidized apo B formation in low-density lipoprotein in human blood. The median inhibitory concentration range was 7–10 µM (*23*). Supplementation of the diet of mice with a dried hexane extract of the leaves (1% w/w of diet) for 1 week reduced the level of phospholipid hydroperoxides in the plasma, red blood cells (65–74% of the levels of non-supplemented control mice) and liver of the animals (*24*).

The antioxidant activities of carnosol and other compounds extracted from rosemary were compared. Carnosol showed potent antioxidative activity, and scavenged 1,1-diphenyl-2-picrylhydrazyl free radicals, as well as protecting DNA in the Fenton reaction (*18*).

An alcohol extract of the crude drug was assessed in vitro and in vivo for its ability to protect against free radical-induced skin damage. The extract inhibited oxidative alterations to skin surface lipids (*25*). The protective effect of a 96% ethanol extract of the crude drug against oxidative DNA damage induced by hydrogen peroxide and visible light-excited methylene blue in colon cancer cells (CaCo-2) and hamster lung cells (V79) was investigated. The level of DNA damage (DNA strand-breaks) was measured using the comet assay. The extract reduced the genotoxic activity of both agents after long-term (24 h; 0.3 µg/ml) or short-term (2 h; 30.0 µg/ml) pre-incubation of cells, demonstrating that the extract

has a protective effect against oxidative damage to DNA as a consequence of scavenging of both hydroxyl radicals and singlet oxygen (*26*).

Antimicrobial activity

A commercial extract of the leaves (no further details of extract given), dissolved in ethanol (100.0 mg/ml) was tested against foodborne microorganisms. In the Gram-positive bacteria, the minimum inhibitory concentration of the ethanolic solution was 1% for *Leuconostoc mesenteroides,* 0.5% for *Listeria monocytogenes,* 0.5% for *Staphylococcus aureus*, 0.13% for *Streptococcus mutans* and 0.06% for *Bacillus cereus*. It was fungistatic in *Penicillium roquefortii* and *Botrytis cinerea*. Up to 1% of the ethanolic solution had no activity against the Gram-negative bacteria *Escherichia coli*, *Salmonella enteritidis* and *Erwinia carotovora* and on the yeasts *Rhodotorula glutinis* and *Cryptococcus laurentii*. The antibacterial activity was associated with phenolic diterpenoids in the hexane extract of the leaves (*27*).

An extract of the leaves inhibited the growth of *Shigella flexneri* and *S. sonnei* with a minimum inhibitory concentration ranging from 0.5 to 1% (w/v) depending on the *Shigella* strain used (*28*).

Antinephrotoxic activity

The effects of rosmarinic acid on the suppression of mesangioproliferative glomerulonephritis, induced by intravenous injection of rabbit anti-rat thymocyte serum to rats was assessed. Intragastric administration of rosmarinic acid to the rats at a dose of 100.0 mg/kg bw per day for 8 days decreased the quantity of proliferating cell nuclear antigen, fibronectin, type IV collagen and fibrin in the glomerulus. Superoxide dismutase activity of renal cortex homogenate was also significantly augmented in animals treated with rosmarinic acid (*29*).

Antitrypanosomal activity

A methanol extract of the leaves at a concentration of 2.0 mg/ml inhibited the motility of cultured epimastigotes of *Trypanosoma cruzi* after 2 h of incubation. Two triterpene acids, oleanolic and ursolic acids, isolated from the extract were responsible for the activity. Ursolic acid stopped the movement of all *T. cruzi* epimastigotes at the minimum inhibitory concentration of 40.0 µg/ml (88.0 µM) after 48 h of incubation. Oleanolic acid was less active, with a minimum inhibitory concentration of 250.0 µg/ml (550.0 µM) (*30*).

Antitumour activity

The effect of dietary intake of an extract of the leaves on 7,12-dimethylbenz[a]anthracene-induced mammary tumorigenesis and on the

in vivo formation of mammary 7,12-dimethylbenz[a]anthracene-DNA adducts was evaluated. Supplementation of a semi-purified diet with 1.0% by weight of the rosemary extract resulted in a significant (47%, $p < 0.01$) decrease in incidence of mammary tumours compared to controls. In subsequent studies, dietary supplementation with 0.5% and 1.0% rosemary extract inhibited total in vivo binding of 7,12-dimethylbenz[a]anthracene to mammary epithelial cell DNA by an average of 42%. This decrease in total binding was not due to a uniform decrease in the formation of all mammary 7,12-dimethylbenz[a]anthracene-DNA adducts (*31*).

Antiulcer activity

An ethanol (70%) extract was evaluated for antiulcerogenic activity in vivo. Intragastric administration of 100.0 mg/kg bw per day to 1.0 g/kg bw per day of the extract decreased the ulcerative lesion index produced by ethanol and reserpine in rats. No antisecretory activity was observed in the pyloric ligation model. Prior administration of L-NAME (N[G]-nitro L-arginine methyl ester), a nitric oxide-synthase inhibitor, did not reduce the antiulcerogenic activity of the extract in the ethanol-induced ulcer model, suggesting that the pharmacological mechanism has no relationship with nitric oxide. When the animal groups were treated with indomethacin using the same experimental model, the extract did not reduce the antiulcerogenic activity, suggesting that the pharmacological mechanism has no relationship with prostaglandins. The previous treatment with N-ethylmaleimide, a thiol blocker, including mucosal nonprotein sulfhydryl groups, reduced the antiulcerogenic activity of the extract in the ethanol-induced ulcer model. This result suggests that the ethanol extract has active substances that increase the content of mucosal nonprotein sulfhydryl groups (*32*).

Diuretic effects

The effects of aqueous extracts of the crude drug on the treatment of kidney function and diuresis in rats were determined. Outcomes assessed included urinary volume and the excretion of sodium, potassium and chloride. The concentration of electrolytes and urea in plasma and creatinine clearance were also measured. Daily intragastric administration of the aqueous extracts of the leaves, at a dose of 10 ml/kg bw of an 8% or 16% extract in distilled water for 1 week, significantly enhanced diuresis in rats compared to the control group from the fifth day of treatment ($p < 0.001$). At a dose of 8%, the peak of urinary excretion of sodium, potassium and chloride was reached after 6 days of treatment ($p < 0.001$). A dose of 16% did not significantly affect the excretion of water and electrolytes over a similar period but significantly increased the urinary ex-

cretion of sodium and chloride on the seventh day and of potassium on the sixth day ($p < 0.05$). No change was observed in plasma electrolytes and urea in any group, except for a decrease in sodium and chloride concentration in the group treated with the 16% extract of the crude drug. A decrease in creatinine clearance was observed after treatment with a daily dose of the 8% extract (*33*).

Enzyme induction

The effects of an aqueous extract of the leaves and of rosmarinic acid were investigated on xenobiotic metabolizing enzymes in rat liver after dietary administration (0.5% daily rations) for 2 weeks. The modulation of phase I enzymes such as cytochrome P450 1A, 2B, 2E1, 3A and phase II enzymes such as glutathione S-transferase, quinone reductase and uridine diphosphate (UDP)-glucuronosyltransferase was evaluated by measuring enzyme activities with specific substrates. The aqueous extract enhanced cytochrome P450 1A1, 2B1/2, 2E1 and glutathione S-transferase (especially recombinant glutathione S-transferase A3/A5, M1 and M2), quinone reductase and UDP-glucuronosyltransferase. No modification of xenobiotic metabolizing enzymes was observed in response to treatment with rosmarinic acid, indicating that other constituents of the leaves are responsible for this activity (*34*).

The ability of the leaves to modulate cytochrome P450 and detoxification enzymes in rat liver was evaluated by comparing the effects of dried leaves and leaf extracts with those of the essential oil. Rats received the powdered leaves or extracts of the leaves in their diet at 0.5% (w/w) for 2 weeks. The effects of such treatments were evaluated for cytochrome P450 isozymes 1A, 2B, 2E1, glutathione S-transferase, NAD(P)H, quinone reductase and UDP-glucuronosyltransferase activities and on protein levels (immunoblot analyses). The results demonstrated that essential oil selectively induced cytochrome P450, particularly cytochrome P450 2B. The leaf extract enhanced both cytochrome P450 and detoxification enzymes. A dichloromethane extract of the leaves acted as a monofunctional inducer, inducing glutathione S-transferase, quinone reductase and UDP-glucuronosyltransferase, in particular UDP-glucuronosyltransferase 1A6 (*35*).

An extract of the crude drug was fed to female mice at concentrations of 0.3% and 0.6% (by weight) for 4 weeks prior to determination of the activities of the detoxification enzymes glutathione-S-transferase and NAD(P)H, and quinone reductase in lung, liver and stomach. Liver activities of glutathione S-transferase and quinone reductase, and stomach glutathione S-transferase activity, were significantly increased in animals fed diets containing the extract. However, diets supplemented with the

extract did not affect lung glutathione S-transferase and quinone reductase activities (*36*).

Estrogenic effects

The effects of a methanol extract of the leaves on the metabolism and action of estradiol and estrone were assessed in vivo. Treatment of female mice with 2% rosemary in an American Institute of Nutrition (AIN)-76A diet for 3 weeks increased the liver microsomal 2-hydroxylation of estradiol and estrone by approximately 150%, increased their 6-hydroxylation by approximately 30% and inhibited the 16α-hydroxylation of estradiol by approximately 50%. Treatment of female CD-1 mice with 2% rosemary in the diet for 3 weeks also stimulated the liver microsomal glucuronidation of estradiol and estrone by 54–67% and 37–56%, respectively. In further studies, feeding 2% rosemary in the diet to ovariectomized CD-1 mice for 3 weeks inhibited the uterotropic action of estradiol and estrone by 35–50% compared with animals fed a control diet (*37*).

Immune stimulant activity

The effect of an ethanol extract of the leaves on splenic mononuclear cell proliferation was determined in vivo. Rats were fed diets containing 0, 100, 200 or 400 ppm leaf extract or 400 ppm butylated hydroxytoluene in combination with 10 or 20% casein-enriched diets for 8 weeks. Splenic mononuclear cells were isolated from these animals and the mitogenic response to concanavalin A, phytohaemagglutinin and lipopolysaccharide was determined. Concanavalin A- and phytohaemagglutinin-stimulated proliferation of spleen cells in rats fed 10% casein and 200 ppm leaf extract was significantly higher than that of cells from the corresponding control animals. Other concentrations of the extract were not active, suggesting that the leaf extract does not have a generalized immune-enhancing effect (*38*).

Rosmarinic acid induced apoptosis in a p56(lck) (Lck)-dependent manner. Lck(+) Jurkat T cells underwent apoptosis in response to treatment with rosmarinic acid , whereas Lck(-) Jurkat subclone J.CaM1.6 cells did not. J.CaM1.6 cells with various Lck mutants indicated that Lck SH2 domain, but not Lck kinase activity, was required for rosmarinic acid-induced apoptosis. Rosmarinic acid-mediated apoptosis involved a mitochondrial pathway as indicated by cytochrome c release and the complete blockage of apoptosis by an inhibitor of mitochondrial membrane depolarization. Both caspase-3 and caspase-8 were involved in rosmarinic acid-induced apoptosis and work downstream of mitochondria and caspase-9 in the order of caspase-9/caspase-3/caspase-8. In freshly isolated human peripheral blood mononuclear cells, rosmarinic acid

specifically induced apoptosis of Lck(+) subsets such as T and NK cells, but not Lck-deficient cells, including B cells and monocytes. Moreover, the ability of rosmarinic acid to kill T and NK cells was restricted to actively proliferating cells, and was not seen in resting cells (*39*).

Toxicology

To find out whether the crude drug induces abortion and/or interferes with the normal development of the concepts, doses of 26 mg of a 30% (w/v) aqueous extract (13 mg solids/ml) made with leaves, flowers and stem, were administered daily to rats by gavage during two different periods of pregnancy. One group of animals (n = 12) received the extract from days 1 to 6 of pregnancy (pre-implantation period) and another group (n = 14) received the same extract from days 6 to 15 of pregnancy (organogenic period). Control groups (n = 12) received saline solution at the same volume and during the same periods as the comparable experimental groups. The animals were killed at term. The treatment of the dams during either the pre-implantation or the organogenic period did not cause significant changes in the post-implantation loss or in the number of anomalies or malformations of the term fetuses, which also showed a similar degree of development to that of the control animals. The percentage of pre-implantation loss in the group treated before embryo implantation increased compared to the control group, although the difference was not statistically significant. This result suggests that rosemary extract may have an anti-implantation effect without interfering with the normal development of the concept after implantation (*40*).

Clinical pharmacology

One of the postulated mechanisms of action of phenolic compounds as antioxidants is chelation of pro-oxidant metals, such as iron. Although the antioxidant activity is viewed as a positive effect, this activity may impair the utilization of dietary iron. A small clinical study assessed the effect of phenolic-rich extracts obtained from green tea or the crude drug on nonhaem-iron absorption. Twenty-four female volunteers consumed test meals on four separate occasions. The meals were identical except for the absence (meal A) or presence (meal B) of a phenolic-rich extract from green tea (study 1; n = 10) or rosemary (study 2; n = 14). The extracts (0.1 mM) were added to the meat component of the test meals. The meals were labelled with either ^{55}Fe or ^{59}Fe and were consumed on four consecutive days in the order ABBA or BAAB. Iron absorption was determined by measuring whole-body retention of ^{59}Fe and the ratio of ^{55}Fe or ^{59}Fe activity in blood samples. The results demonstrated that the presence of phenolic-rich extracts resulted in decreased non-haem-iron absorption.

Mean (± standard deviation) iron absorption decreased from 12.1 ± 4.5% to 8.9 ± 5.2% ($p < 0.01$) in the presence of green tea extract and from 7.5 ± 4.0% to 6.4 ± 4.7% ($p < 0.05$) in the presence of the rosemary extract (*41*).

Adverse reactions

There is a single case-report of photoaggravated allergic contact dermatitis, in which a patient developed contact dermatitis after handling the leaves of the plant on a sunny day (*42–44*). One case of cheilitis has been reported (*45*). A 56-year-old man developed occupational contact dermatitis of his hands, forearms, and face after coming into contact with an extract of the leaves. He reacted to carnosol, the main constituent of the extract (*44*).

Contraindications

Folium Rosmarini is contraindicated in cases of hypersensitivity or allergy to the plant material.

Warnings

No information was found.

Precautions

Drug interactions

While no drug interactions have been reported, an aqueous extract of the crude drug enhanced the activity of cytochrome P450 1A1, 2B1/2, 2E1 and glutathione S-transferase (especially recombinant glutathione S-transferase A3/A5, M1 and M2), quinone reductase and UDP-glucuronosyltransferase (*34*). Thus, drugs metabolized through these cytochrome P450 isozymes may be affected.

Carcinogenesis, mutagenesis, impairment of fertility

An ethanol extract of the leaves was not mutagenic in the Hist revertant *Salmonella typhimurium* TA 1530 strain at a concentration of 5 µg/ml (*46*). See also the study by Lemonica (*40*) under Toxicology.

Pregnancy: non-teratogenic effects

See Toxicology.

Other precautions

No information was found.

Dosage forms

Crude drug for infusions, dry extracts, fluidextract and other Galenical preparations for internal and external use (*47*).

Posology

(Unless otherwise specified)
Daily dosage: for oral use 4–6 g of herb. Infusion: 2–4 g in 150 ml water three times daily. Fluidextract (1:1, 45% ethanol w/w) 1.5–3.0 ml daily. Tincture (1:5, 70% ethanol) 3–8.5 ml daily. Dry extract (4.5–5.5:1 w/w) 0.36–0.44 g, three times daily. External use: boil 50 g of herb in 1 l of water, add to one full bath (*47*).

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Cortex Salicis

Definition
Cortex Salicis consists of the whole or fragmented dried bark from young branches of *Salix alba* L., *S. daphnoides* Vill., *S. fragilis* L., *S. purpurea* L., and other appropriate *Salix* species (Salicaceae) (*1–4*).

Synonyms
No information was found.

Selected vernacular names
Salix alba L.: Ak sõyüd ag, basket willow, bela vrba, beli, bid-e-maamouli, caporniolo, derakht-e-bid, European willow, hopeapaju, hvid pil, isbîdâr, kvitpil, osier blanc, paju, remmelgas, salcio bianco, salicastro, salcio da forche, salece, salgueiro-de-casa-roxa, saligastro, sargatillo, saule blanc, sauce blanco, Silberweide, sogut, solvpil, sufsaf abiad, tortiello, swallow tailed willow, vitpil, white willow (*3*, *5–10*).

Salix daphnoides Vill.: Daphne willow, Reiweide, salicio nero, saule à bois glauque, saule faux daphné, saule noir, Schimmel Weide, vi, violet willow, wierzba wawrzynkolistna (*3*, *10*).

Salix fragilis L.: Bid-khesht, brittle willow, Bruckweide, common crack willow, crack willow, Éva-fuz, hrökkvíòir, krhka vrba, Krackweide, kraakwilg, piilipuu, rabe remmelgas, salice, saliva, saule fragile, skjorpil, skörpil, sufsaf, törékeny fuz (*3*, *5*, *8*, *10*).

Salix purpurea L.: Bidsorkh, morvár, osier rouge, purple osier, purple willow, purpur pil, Pupurweide, salcio da vimini, salcio rosso, sorkhbid, sauce Colorado, saule pourpre, wierzba purpurowa (*3*, *5*, *10*).

Geographical distribution
Native to Europe and Asia, and naturalized in North America (*6*, *11*).

Description
Salix species are woody trees, up to 25 m in height. Leaves are deciduous, generally petiolate and stipulate, simple, alternate to subopposite, linear

to widely obovate, margins entire to serrulate. Leaf underside is generally hairy or glaucous, rarely glabrous. Inflorescence consists of small unisexual catkins emerging, depending on species, either before, at the same time as, or after the leaves emerge; individual flowers inconspicuous, apetalous, subtended by a single fringed or hairy bract; sepals replaced by nectaries. Stamens 1–8, often 2 in staminate flower, and in pistillate flowers, the ovary is superior, bicarpellate, unilocular; style 1; stigmas 2, each 0–2-lobed. Fruit capsule, two-valved. Seeds many, comose (*1*). *Salix alba* is a large tree with a short trunk, yellowish-brown branches and elliptic-lanceolate, acuminate and serrulate, ash-grey, sericious leaves. The fruit is a capsule dehiscent by 2 valves and contains numerous seeds, on each of which is a basal tuft of hair (*11*).

Plant material of interest: dried branch bark

General appearance

The bark is 1–2 cm wide and 1–2 mm thick and occurs in flexible, elongated, quilled or curved pieces. The outer surface is smooth or slightly wrinkled longitudinally and greenish-yellow in the younger bark to brownish-grey in the older bark. The inner surface is smooth or finely striated longitudinally and white, pale yellow or reddish-brown, depending on the species. The fracture is short in the outer part and coarsely fibrous in the inner region, and is easily split longitudinally. The diameter of current year twigs is not more than 10 mm. The wood is white or pale yellow (*2*, *4*).

Organoleptic properties

Odour: slight; taste: astringent and bitter (*1*, *2*, *4*).

Microscopic characteristics

Two or three rows of poorly developed cork cells with thickened outer walls; cortex of collenchymatous and parenchymatous cells. The latter contain cluster crystals of calcium oxalate, 20–25 µm in diameter and occasionally tannin. Phloem is characterized by tangential groups of lignified fibres associated with a crystal sheath containing prismatic crystals of calcium oxalate. Simple, rounded starch granules 6–8 µm in diameter in the parenchymatous cells of the phloem and medullary rays (*2*).

Powdered plant material

Pale yellow, greenish-yellow or light brown. Microscopically, bundles of narrow fibres, up to about 600 µm long, with very thick walls, lignified, and surrounded by a crystal sheath containing prism crystals of calcium oxalate; parenchyma of the cortex with thick, pitted and deeply beaded walls, and containing large cluster crystals of calcium oxalate; uniseriate

medullary rays; thickened and suberized cork cells. Groups of brownish collenchyma from the bud may be present. Twigs show, additionally, fragments of lignified fibres and vessels from the xylem (*2*, *4*).

General identity tests

Macroscopic and microscopic examinations and thin-layer chromatography (*1*, *2*, *4*).

Purity tests

Microbiological

Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*12*).

Foreign organic matter

Not more than 3% of twigs with a diameter greater than 10 mm, and not more than 2% other foreign matter (*1*, *2*, *4*).

Total ash

Not more than 10% (*1*, *2*, *4*).

Acid-insoluble ash

Not more than 3% (*1*, *2*).

Water-soluble extractive

Not less than 10% (*1*, *2*).

Alcohol-soluble extractive

To be established in accordance with national requirements.

Loss on drying

Not more than 11% (*4*).

Pesticide residues

The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*4*). For other pesticides, see the *European pharmacopoeia* (*4*) and the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*12*) and pesticide residues (*13*).

Heavy metals

For maximum limits and analysis of heavy metals, consult the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*12*).

Radioactive residues

Where applicable, consult the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*12*).

Chemical assays

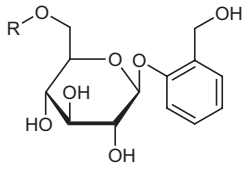
Not less than 1.5% of total salicylate derivatives expressed as salicin by high-performance liquid chromatography (*4*).

Major chemical constituents

The major biologically active constituents are the phenolic glycosides including salicin (approximately 1%), salicortin (up to 4.0%), 2´-*O*-acetylsalicortin (up to 10%), 2´-*O*-acetylsalicin (= fragilin, up to 4%), tremulacin (0.12–2%), 3´- and 4´-acetylsalicortin, populin and salireposide, which have collectively been designated as "salicylates" (*6*). Triandrin, vimalin, picein and grandidentatin are non-saligenin structure-based phenolic compounds. Other significant constituents are flavonoids and tannins (*1*, *5*, *6*).

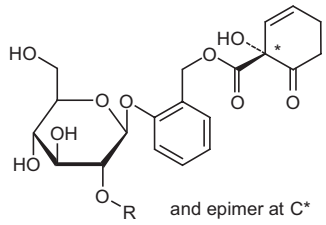
Total salicin content (after hydrolysis) varies according to species. Species rich in total salicin include *S. daphnoides* (2–10%), *S. purpurea* (4–8.5%), *S. fragilis* (2–10%) and *S. alba* (0.5–1%) (*1*, *6*).

The structures of salicin, salicortin, 2´-*O*-acetylsalicortin and 6´-*O*-acetylsalicin (= fragilin) are presented below.



Salicin R = H

Fragilin R = CO-CH_3



and epimer at C*

Salicortin
R = H

2'-*O*-Acetylsalicortin
R = CO-CH_3

Medicinal uses

Uses supported by clinical data

Used orally for the symptomatic treatment of fever and pain, and symptomatic treatment of mild rheumatic conditions (*14–20*).

Uses described in pharmacopoeias and well established documents

Used orally for the treatment of the common cold (*1*).

Uses described in traditional medicine

Used orally for the treatment of constipation and urinary incontinence. Used externally for the treatment of warts (*5*).

Pharmacology

Experimental pharmacology

Anti-inflammatory activity

Similar to salicylates, such as acetylsalicylic acid, extracts of the crude drug are thought to act by inhibiting the activity of cyclooxygenase and thereby inhibiting the synthesis of prostaglandins, which play a role in inflammation, fever and pain. Acetylsalicylic acid inhibits the synthesis of prostaglandins through the acetylation of the enzyme cyclooxygenase. Salicylic acid, and the salicylates that lack an acetyl group, reduce prostaglandin synthesis via inhibition of the activity of cyclooxygenase II (*1*, *21*).

Intragastric administration of 100.0 mg/kg body weight (bw) of tremulacin, a constituent of the bark, significantly inhibited carrageenan-induced hind paw oedema in rats ($p < 0.001$) (*22*). Intragastric administration of 0.5 ml/kg bw of a 30% ethanol extract of the bark to rats reduced carrageenan-induced hind paw oedema, but was not effective in the adjuvant-induced arthritis or dextran-induced pedal oedema models (*23*).

Antipyretic activity

Intragastric administration of 0.8 ml/kg bw of a 30% ethanol extract to rats, suppressed yeast-induced pyrexia (*23*). The antipyretic effects of salicin, saligenin (an aglycone of salicin) and salicylic acid (an active metabolite of salicin) were assessed in rats (*24*). After intragastric administration of salicin to rats, the metabolite salicylic acid appeared slowly in the plasma and concentrations increased gradually, in contrast to the rapid appearance observed after oral administration of sodium salicylate or saligenin. At a dose of 5 mM/kg bw, orally administered salicin did not affect the rectal temperature of afebrile rats while at the same dose, sodium salicylate and saligenin lowered body temperature significantly. However, salicin significantly reduced yeast-induced fever ($p < 0.01$), producing a normal body temperature, and completely prevented fever when administered simultaneously with yeast. Salicin did not induce gastric lesions even at a dose of 5 mM/kg bw; conversely, sodium salicylate and saligenin induced severe gastric lesions in a dose-dependent manner when administered at doses of 1, 2.5 and 5 mM/kg bw (*24*).

Pharmacokinetics

The pharmacokinetics of salicin, saligenin (an aglycone of salicin) and salicylic acid (an active metabolite of salicin) were determined in rats (*24*). Poor absorption of salicin and rapid absorption of salicylic acid and saligenin were confirmed in this animal model. Only small amounts of salicylic acid and saligenin were detected in the intestinal tracts of rats 1 h

after oral administration. More than 50% of a salicin dose was recovered as salicin and saligenin from the intestinal tracts 1 h after treatment and 15.8% of the dose was still present as saligenin 4 h after administration. When given to germ-free rats, 19.8% of the salicin dose was recovered intact, mainly from the cecum, and no saligenin was detected even at 4 h after treatment. These results indicate that salicin is a pro-drug that is gradually transported to the lower part of the intestine, hydrolysed to saligenin by intestinal bacteria, and converted to salicylic acid after absorption. It thus produces an antipyretic action without causing gastric injury (*25*).

Toxicology

Intragastric administration of a 40% ethanol extract of the bark to rats at a dose of 1.6 ml/kg bw for 13 weeks had no effect on kidney function, haematological parameters, liver function or cholesterol levels (*26*). Histological evaluation of the animals after killing showed no pathological changes in the brain, heart, lungs, bone, kidneys, liver, reproductive organs, mammary tissue, stomach or intestines (*26*).

The median lethal dose range of a 30% ethanol extract of the bark was 28.0–42.0 ml/kg bw in mice (both sexes) (*26*).

Clinical pharmacology

Anticoagulant activity

A study was performed to investigate the anticoagulant activity of a bark extract used in the treatment of 51 patients with chronic back pain. Thirty-five patients suffering from acute exacerbations of chronic low back pain received randomly and double-blind either the extract (corresponding to 240 mg salicin/day; $n = 19$) or a placebo ($n = 16$). A further 16 patients with stable chronic ischaemic heart disease were given 100 mg acetylsalicylate per day. Platelet aggregation was studied after drawing blood from the treated patients using an aggregometer. Arachidonic acid (500 µg/ml), adenosine diphosphate (2×10^{-5} M) and collagen (0.18 µg/ml) were used as aggregating agents. The mean maximal arachidonic acid-induced platelet aggregation was 61%, 78% and 13% for the groups treated with extract, placebo and acetylsalicylate, respectively. Acetylsalicylate had a significant inhibitory effect on platelet aggregation compared to the extract ($p = 0.001$) and placebo ($p = 0.001$). There was also a significant difference between the placebo and the extract-treated groups in the maximal platelet aggregation induced by arachidonic acid ($p = 0.04$) and ADP ($p = 0.01$). No statistical difference was found between platelet aggregation in the three groups when collagen was added to the human platelets.

Daily consumption of the extract with 240.0 mg salicin per day affected platelet aggregation to a lesser extent than acetylsalicylate (*27*).

Chronic lower back pain

Standardized extracts of the bark exhibited analgesic effects similar to those of higher doses of acetylsalicylic acid. A daily dose of 1572.0 mg of an extract of the crude drug (standardized to 15.2% salicin or 240.0 mg salicin per day) was superior to placebo in the treatment of pain in patients with osteoarthritis of the hip and the knee, and in patients with exacerbations of chronic low back pain. In two open studies, unspecified extracts of the crude drug exhibited similar efficacy to the normal treatment regime of nonsteroidal antirheumatic drugs and the efficacy was similar to that of refecoxib (*18*).

A randomized placebo-controlled clinical study evaluated the efficacy of a willow bark extract in 210 patients with chronic lower back pain. The patients were assigned to receive an orally administered willow bark extract with either 120.0 mg (low dose) or 240.0 mg (high dose) of salicin, or a placebo, with tramadol as the sole rescue medication, in the 4-week trial. The principal outcome measure was the proportion of patients who were pain-free without tramadol for at least 5 days during the final week of the study. A total of 191 patients completed the study. The number of pain-free patients in the last week of treatment was 27 (39%) of 65 in the group receiving high-dose extract, 15 (21%) of 67 in the group receiving low-dose extract, and 4 (6%) of 59 in the group treated with placebo ($p < 0.001$). The response in the group treated with the high dose was evident after only 1 week of treatment. Significantly more patients in the group receiving the placebo required tramadol during each week of the study ($p < 0.001$). One patient in the treatment group suffered a severe allergic reaction (*14*).

An open, non-randomized study (postmarketing surveillance) assessed the efficacy of an extract of the crude drug in three groups of patients aged 18–80 years presenting over an 18-month period with acute exacerbations of low back pain. The first group of 115 patients was prescribed a daily dose of the extract containing 120.0 mg of salicin. A second group of 112 patients was prescribed the extract equivalent to 240.0 mg salicin per day. A third "control" or "comparator" group of 224 patients received conventional therapy. In the patients who had received conventional therapy, the exacerbations had been shorter but the pain was more intense as judged by Arhus Index and Total Pain Index. After 4 weeks of treatment, about 40% of patients in the group treated with the extract equivalent to 240 mg salicin per day were free of pain. In the group treated with extract containing 120.0 mg of salicin, as a whole, about 19% of patients were

pain-free at 4 weeks, but only 8% of those who did not resort to supplementary treatment. In the group receiving conventional therapy, 18% of patients were pain-free. Better pain relief in the group treated with the equivalent of 240.0 mg salicin was accompanied by a reduction in the use of supplementary conventional treatments (*15*).

Migraine

A placebo-controlled double-blind clinical trial involving 54 patients assessed the efficacy of a topical medication containing salicin for the treatment of either migraines and/or tension-type headaches. All the patients had headaches consistent with the International Classification of Headache Disorders (HIS) criteria for migraines and/or tension-type headaches, and had suffered from headaches for at least 1 year up to approximately 40 years. The patients were divided into two groups. One group received a placebo topical medication. The other group received a topical medication containing salicin. Neither preparation induced any stinging sensation. The patients were instructed to apply the medication, which was in a roll-on form, to the frontal region at the onset of a headache, take their usual oral or parenteral medications, apply a mask which covered the eyes and frontal region, and then lie down. They subsequently filled out forms rating the degree of relief which they attributed to the topical medications and the masks using a scale of 0 to 10. They were also asked if this form of treatment had helped or not. Seven of the 20 patients who received the placebo stated that the medication and mask helped. This group rated the treatment an average of 4.31 on the scale of 0 to 10. Twenty-eight of 34 patients who received salicin stated that it was effective. This group rated it 7.42 on the scale of 0 to 10 ($p < 0.001$). The majority of the patients treated with salicin stated that the duration of their headaches was significantly reduced, as was their need for analgesic and/or narcotic medications. This study demonstrates a significant difference between placebo and salicin in association with the photoprotective mask in treating migraines and/or tension-type headaches with associated frontalis pain and photophobia (*17*).

Osteoarthritis

A clinical study involving 78 patients assessed the efficacy of a chemically standardized willow bark extract in the treatment of osteoarthritis. Willow bark extract, at a dose corresponding to 240.0 mg salicin/day, was compared to a placebo in a 2-week, double-blind, randomized controlled trial. The primary outcome measure was the pain dimension of the Western Ontario and McMaster Universities (WOMAC) Osteoarthritis Index. Secondary outcome measures included the stiffness and physical

function dimensions of the WOMAC, daily visual analogue scales (VAS) on pain and physical function, and final overall assessments by both patients and investigators. The results demonstrated a statistically significant difference between the subjects who received active treatment and the placebo group in the WOMAC pain dimension (d = 6.5 mm, 95% confidence intervals (CI), 0.2–12.7 mm, $p = 0.047$); the WOMAC pain score was reduced by 14% from the baseline level after 2 weeks of active treatment, whereas there was an increase of 2% in the placebo group. The patient diary VAS confirmed the results, and the final overall assessments showed superiority of the willow bark extract over the placebo (patients' assessment, $p = 0.0002$; investigators' assessment, $p = 0.0073$) (*19*, *20*).

A double-blind, randomized, controlled clinical trial assessed the efficacy of an extract of the bark (containing 17.6% total salicin), at a dose of 240.0 mg per day, in the treatment of patients with chronic arthritic pain. Eighty-two subjects with chronic arthritic pain were randomly assigned for 2 months without cross-over to receive either the extract or a placebo. The results demonstrated a small but statistically significant improvement in pain symptoms ($p < 0.05$), although the improvement was less in patients with osteoarthritis. No other significant changes in any other measures or in the use of other self-prescribed analgesics were observed (*28*).

The efficacy of a standardized willow bark extract was investigated in two randomized, controlled, double-blind trials with follow-up for 6 weeks, in patients with osteoarthritis and rheumatoid arthritis. One hundred and twenty-seven outpatients with osteoarthritis of the hip or knee and a pain score of at least 30 mm, and 26 outpatients with active rheumatoid arthritis were randomly allocated to one of three groups. Patients in the first group received willow bark extract, corresponding to 240 mg of salicin/day; patients in the second group, diclofenac at a dose of 100 mg/day; and the third group, a placebo ($n = 43$, 43 and 41, respectively). The main outcome measure was the pain subscore. In the rheumatoid arthritis trial, patients were randomly assigned to receive either willow bark extract, corresponding to 240 mg salicin/day ($n = 13$) or a placebo ($n = 13$). The main outcome measure was the patient's assessment of pain rated on a 100-mm visual analogue scale. In the osteoarthritis trial, pain scores decreased by 8 mm (17%) in the group treated with willow bark and by 23 mm (47%) in the group treated with diclofenac, compared with 5 mm (10%) in the patients who received the placebo. The difference between the scores following treatment with willow bark extract and treatment with placebo was not statistically significant (–2.8 mm; 95% CI –12.1 to 6.4 mm; $p = 0.55$, analysis of covariance (ANCOVA)), but the difference between the outcomes of treatment with diclofenac and with

the placebo was highly significant (–18.0 mm; 95% CI –27.2 to –8.8 mm; p = 0.0002, ANCOVA). The results in the rheumatoid arthritis trial showed that the mean reduction of pain reported using the visual analogue scale was –8 mm (15%) in the group treated with willow bark compared with –2 mm (4%) in the placebo group. The difference was not statistically significant (estimated difference –0.8 mm; 95% CI –20.9 to 19.3 mm; p = 0.93, ANCOVA) (*29*).

Pharmacokinetics

The pharmacokinetics of salicin and its major metabolites were determined in humans after oral administration of an extract of the crude drug. The extract, corresponding to 240.0 mg salicin (1360 mg, 838 µmol), was administered to 10 healthy volunteers in two equal doses at times 0 h and 3 h. Over a period of 24 h, levels of salicylic acid and its metabolites in urine and serum, i.e. gentisic acid and salicyluric acid, were determined using reverse-phase high-performance liquid chromatography. Renal excretion rate, elimination half-life and total bioavailability of salicylates were calculated. The results showed that salicylic acid was the major metabolite of salicin detected in the serum (86% of total salicylates), as well as salicyluric acid (10%) and gentisic acid (4%). Peak serum levels were reached within less than 2 h after oral administration. Renal elimination occurred predominantly in the form of salicyluric acid. Peak serum levels of salicylic acid were on average 1.2 mg/l, and the observed area under the serum concentration–time curve of salicylic acid was equivalent to that expected from an intake of 87.0 mg acetylsalicylic acid (*30*).

Adverse reactions

Allergic reactions such as pruritus, urticaria, asthma and gastrointestinal symptoms may occur (*16*). One case of an allergic reaction in a 32-year-old atopic patient who showed a severe anaphylactic reaction after the ingestion of a pollen compound containing the crude drug has been reported (*31*). One case of anaphylaxis resulting from the use of an extract of the crude drug in a patient with a history of an aspirin allergy has been reported (*32*).

Contraindications

Cortex Salicis is contraindicated in cases of hypersensitivity or allergy to the plant material or to salicylates (e.g. asthma, bronchial spasm, rhinitis or urticaria).

It is also contraindicated during pregnancy and lactation, in patients with salicylate intolerance and patients with impaired thrombocyte function (*7*, *27*), and in children under the age of 12 years (*7*).

Warnings

In children under the age of 12 years, Cortex Salicis should only be used on the advice of a health care professional due to the possibility of Reye's syndrome. In cases of a child or adolescent who has become very ill with severe vomiting, drowsiness or loss of consciousness following a viral infection, Reye's syndrome should be suspected. This extremely rare, life-threatening disease requires immediate medical attention.

In cases of severe liver or renal dysfunction, coagulation disorders, gastric/duodenal ulcer and glucose-6-phosphate dehydrogenase deficiency, the product should only be taken under medical supervision.

Consult a health care professional in cases of fever (> 39 °C), lasting longer than 3 days. A health care professional should also be consulted if acute conditions of swelling of joints, redness and impaired mobility persist or worsen during the first week of use of Cortex Salicis.

Precautions

General

The use of Cortex Salicis in patients with hypersensitivity to other non-steroidal anti-inflammatory drugs is not recommended. Use should be avoided in people with asthma because severe reactions (acute bronchospasms) could be induced (*16*).

Drug interactions

Currently there are no data indicating that the crude drug interacts with anticoagulant drugs such as coumarin and heparin. However, Cortex Salicis may increase the effects of anticoagulants. Patients on anticoagulant therapy should not use products containing the crude drug without the supervision of a health care professional (*7*).

Carcinogenesis, mutagenesis, impairment of fertility

Oral administration of a 40% ethanol extract of the bark did not disrupt the estrous cycle, or inhibit ovulation or fertility when administered to female rats and rabbits at a dose of 1.6 ml/kg bw (*26*).

Pregnancy: teratogenic effects

Oral administration of a 40% ethanol extract of the bark to female rats and rabbits at a dose of 1.6 ml/kg bw during pregnancy was not teratogenic (*26*).

Pregnancy: non-teratogenic effects

See Contraindications.

Oral administration of a 40% ethanol extract of the bark was not embryotoxic when administered to rats and rabbits during pregnancy at a dose of 1.6 ml/kg bw (*26*).

Nursing mothers
See Contraindications.
Salicylates cross the placenta and appear in breast milk (*1*).

Paediatric use
See Contraindications.

Other precautions
No information was found.

Dosage forms
Crude drug, dried hydroalcoholic or aqueous extracts, tinctures and fluidextracts (*1*, *16*, *33*).

Posology
(Unless otherwise indicated)
Adult oral daily dose: extracts, tinctures or fluidextracts equivalent to 120–240 mg of total salicin, or 6–12 g of powdered drug material as a decoction (corresponding to 120–240 mg of total salicin) in two divided doses (*1*, *16*, *33*).

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Fructus Tribuli

Definition
Fructus Tribuli consists of the dried fruits of *Tribulus terrestris* L. (Zygophyllaceae) (*1–4*).

Synonyms
Tribulus lanuginosus L. (*5*).

Selected vernacular names
Abrojo, abrojos, akanti, alaf-e-kanguereh, baijili, bastitaj, be tha gokharu, bethu, bhakhra, bullhead, burnut, burra gookeron, calthrop, caltrap, caltrop, cat's head, chotagokhru, cows hoof, croix de chevalier, croix de malte, dars el agûz, dava-tehokourtdi, demirdiken, deshi gokhru, devil's thorn, ekanty, eskrundki, espáákh, espigón, gai ma duong, gatha, ghota, goathead, gokharu gokhru, gokhur, gokshura, goksuraka, gokhri, gokhurkata, gokhyura, gukhura, hamabishi, herbe terrestre, ikshugandha, jili, jilisi, kandaai, khar-e-khasak khurd, khárk-tehârouk khárk-tehârpar, khárkhassak, khôrbâr, khokkrasun, kouleh-tighak, krunda, land caltrops, malteserkors, meetha gokhru, Mexican sanbur, michirkand, mithagokharu, mithogokharu, naalkhar, naam din, nana gokharu, nature's viagara, neggilamullu, neggilu, nerenchi, nerinjil, nerunjil, ookharu pakhda, outb, pakhda, pakhra, palleru, palleru kaya, palleruveru, pedda palgeru, punctur vine, qutiba, saligot terrestre, sannanaggilu, sannaneggilu, sarang, sarate, sekal-tali, sharwandi, sher sher, shitsurishi, small caltrop, svadamstraa, tahkandi, tat le, Teufelsdorm, Texas sandbur, traikantaka, tribolo commune, trikanta, tribule couché, tribule terrestre, tsi li, vejtidsel, zama (*4–10*).

Geographical distribution
Native to the Mediterranean regions, but also found throughout the world (*6*).

Description
A perennial, decumbent, pubescent herb, branches 0.5–1.8 m long, tips ascending; longer leaves up to 7 cm long, with 6–9 pairs of leaflets, shorter

leaves up to 6 cm long with 3–5 pairs of leaflets, midrib ending in a mucro, 0.5–2 mm long; leaflets 6–22 mm by 2.5–9 mm, base obliquely rounded, apex subacute, both surfaces silky, subsessile, stipules falcate, 3–6 mm long, acuminate; pedicel 2–4 cm long, sepals narrowly lanceolate, 7–11 mm long, apex acute, appressed hairy, caducous, petals obovate-cuneate, 1–2 cm by 1–1.5 cm, apex broadly rounded-truncate, bright yellow, stamens subequal, anthers 1 mm long; cocci 4–5, spines stout, sharp, 2 lateral ones largest, pericarp rather thick, corky, seeds 1–3 in each mericarp (*6*).

Plant material of interest: dried fruits

General appearance

A pentagonal fruit 7–12 mm in diameter consists of 5 mericarps, radially arranged. Each mericarp often splitting into single hatchet-shaped, 3–6 mm long mericarp; dorsal surface yellowish-green, prominent, with longitudinal ribs and numerous spinelets, bearing symmetrically a pair of long spines and a pair of short spines; two lateral surfaces rough, with reticular striations, greyish-white. Texture hard. Each mericarp contains 1–3 seeds (*1–3*).

Organoleptic properties

Odourless; taste: bitter and pungent (*1*, *2*).

Microscopic characteristics

Transverse section of fruit shows small epidermal cells of each coccus rectangular; unicellular trichomes in abundance; rilesocarp 6–10 layers of large parenchymatous cells, rosette of calcium oxalate crystals abundant; mesocarp followed by 3–4 compact layers of small cells containing prismatic crystals of calcium oxalate (*4*).

Powdered plant material

Yellow-green. The fibres are lignified, with upper and lower layers in a criss-cross arrangement, a few scattered singly, sometimes fibre bundles connected with stone cells. Stone cells elongated-elliptical or subrounded, occurring in groups. Testa cells polygonal or subsquare, about 30 µm in diameter, walls reticulately thickened, lignified. Prisms of calcium oxalate 8–20 µm in diameter (*1*).

General identity tests

Macroscopic and microscopic examinations (*1*, *2*, *4*), thin-layer chromatography (*2*) and microchemical test (*3*).

Purity tests

Microbiological

Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*11*).

Foreign organic matter

Not more than 4.0% fruit stalks and not more than 1% other foreign matter (*2*, *3*).

Total ash

Not more than 12% (*1*).

Acid-insoluble ash

Not more than 1.5% (*2*).

Water-soluble extractive

Not less than 10% (*4*).

Alcohol-soluble extractive

Not less than 8.5% (*2*).

Water

Not more than 9% (*1*).

Loss on drying

Not less than 11% (*2*).

Pesticide residues

The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*12*). For other pesticides, see the *European pharmacopoeia* (*12*) and the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*11*) and pesticide residues (*13*).

Heavy metals

For maximum limits and analysis of heavy metals, consult the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*11*).

Radioactive residues

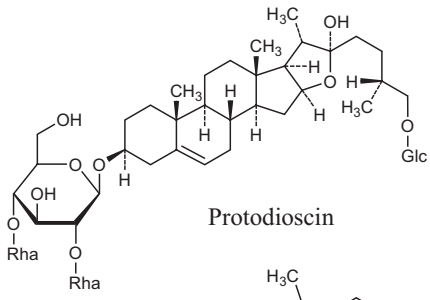
Where applicable, consult the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*11*).

Chemical assays

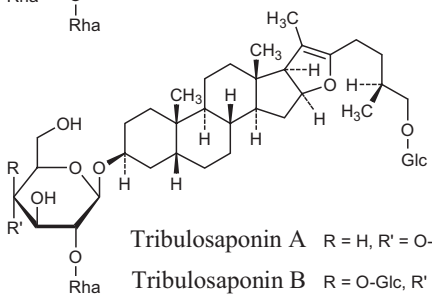
To be established in accordance with national requirements.

Major chemical constituents

The major constituents of the fruit are steroidal saponins including gitonin, protodioscin (0.245%, tribulosaponins A and B, tribulosin and terrestrosins A–K, among others. Other constituents include alkaloids, tribulusamides A and B, and trace amounts of harman and norharman; and flavonols such as kaempferol, quercetin and rutin (*5*, *14*, *15*). The structures of the major chemical constituents are presented below.

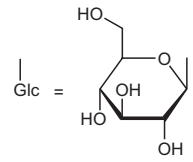


Protodioscin

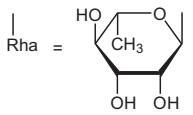


Tribulosaponin A R = H, R' = O-Rha

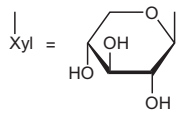
Tribulosaponin B R = O-Glc, R' = H



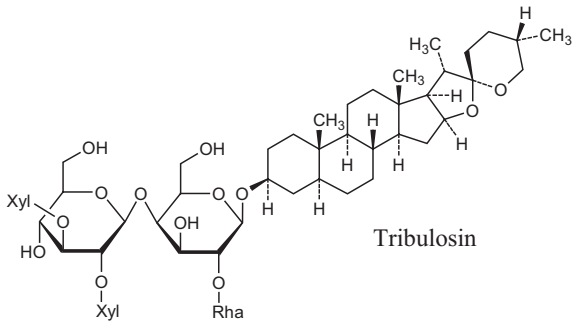
β-D-glucopyranosyl



α-L-rhamnopyranosyl



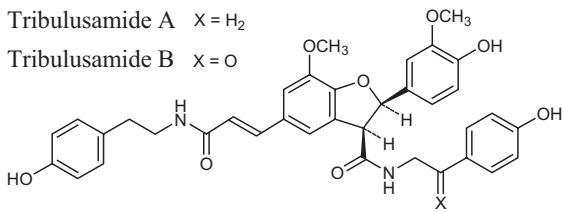
β-D-xylopyranosyl



Tribulosin

Tribulusamide A X = H_2

Tribulusamide B X = O



Medicinal uses

Uses supported by clinical data

None.

Uses described in pharmacopoeias and well established documents

Orally for the treatment of cough, headache and mastitis (*1*).

Although clinical trials have assessed the use of the crude drug for the symptomatic treatment of angina pectoris and male infertility (*16–22*), randomized controlled clinical trials are needed before the use of the crude drug can be recommended for the treatment of these conditions.

Uses described in traditional medicine

Orally for the treatment of abdominal distension, diarrhoea, kidney stones, nosebleeds and vitiligo (*5*, *23*). Also used as an aphrodisiac, diuretic, galactagogue, general tonic and uterine tonic (*5*).

Pharmacology

Experimental pharmacology

Angiotensin-converting enzyme inhibitory effects

The renin-angiotensin-aldosterone system, including angiotensin-converting enzyme plays an important role in the control of blood pressure and fluid volume (*24*, *25*). An aqueous extract of the fruit (1 g fruit in 10 ml water) inhibited the activity of angiotensin-converting enzyme in vitro at a concentration of 0.33 mg/ml (*24*). The mechanism of action of the aqueous extract (500 mg fruit in water 2:1 w/v) was investigated in 2K1C hypertensive rats by measurement of circulatory and local angiotensin converting enzyme activity in aorta, heart, kidney and lung. Hypertension was induced using a silver clip on the renal artery inserted by surgery. The animals were treated with a single oral daily dose of 10 mg/kg body weight (bw) of a lyophilized aqueous extract of the fruit for 4 weeks. The systolic blood pressure was increased in 2K1C rats as compared with control rats. The systolic blood pressure of rats treated with the crude drug extract was significantly decreased compared to hypertensive rats ($p < 0.05$). The angiotensin-converting enzyme activity in all tissues of 2K1C rats including aorta, heart, kidney and lung as well as in the serum was significantly increased compared to that of normal rats. The angiotensin-converting enzyme activity in all tissues of hypertensive rats treated with the fruit extract was significantly lower than that of control hypertensive rats (*26*).

Anti-inflammatory activity
A methanol extract of the fruit inhibited cyclooxygenase 2 activity in lipopolysaccharide-induced mouse macrophages RAW264.7 by > 80% at a concentration of 10.0 µg/ml in vitro (*27*).

Diuretic activity
An aqueous extract of the fruit was dissolved in physiological saline and administered by gavage to rats to determine its diuretic effects. When administered at an intragastric dose of 5.0 g/kg bw, the extract induced diuresis, and was slightly more effective than furosemide. The sodium, potassium and chloride ion concentrations in the urine of animals treated with the extract were also increased. Addition of the extract to the bath medium for strips of isolated guinea-pig ileum induced contractile activity of guinea-pig ileum (*28*).

A dried ether extract of the fruit induced diuresis and increased creatinine clearance in anaesthetized dogs; however no further details of this study were available (*6*).

Hepatoprotective activity
Addition of the tribulusamides A and B, lignanamides isolated from a 70% ethanol extract of the fruit, to a medium containing primary cultured mouse hepatocytes at a concentration of 20 µM significantly ($p < 0.05$) prevented D-galactosamine/tumour necrosis factor alpha-induced cytotoxicity (*14*).

Stimulation of melanocyte proliferation
An aqueous fruit extract was tested in an assay using sulforhodamine B protein stain for cell number to determine if the extract was capable of stimulating melanocyte proliferation, and could be useful for the treatment of vitiligo. Significant stimulation ($p < 0.05$) of melanocyte proliferation was observed, in the absence of tetradecanoyl phorbol acetate, using aqueous extracts of the fruit at concentrations of 0.01–1 mg/ml (*29*).

Reproductive effects
The effect of an extract of the fruit on the isolated corpus cavernosal tissue of New Zealand white rabbits was studied. Twenty-four rabbits were randomly assigned to one of four experimental groups of six animals each. Group 1 served as control. Groups 2, 3 and 4 were treated with an oral dose of the extract at doses of 2.5 mg/kg bw, 5.0 mg/kg bw and 10.0 mg/kg bw, respectively, for 8 weeks. After killing the rabbits, the penile tissue was isolated and evaluated by the response to both contracting and relaxing pharmacological agents and electrical field stimulation. The relaxant responses to electrical field stimulation, acetylcholine and nitroglycerin in

noradrenaline-precontracted corpus cavernosal strips from treated groups showed an increase in relaxation of a concentration-dependent nature when compared to that of the tissues from the control group. However, the contractile, anti-erectile response of corpus cavernosal tissue to noradrenaline and histamine did not differ significantly between the treatment and the control groups. The relaxant responses to acetylcholine, nitroglycerin and electrical field stimulation by more than 10%, 24% and 10%, at the doses of 2.5 mg/kg bw, 5.0 mg/kg bw and 10.0 mg/kg bw, respectively, compared to their control values and the lack of such effect on the contractile response to noradrenaline and histamine indicate that the extract has a pro-erectile activity (*30*).

An extract of the fruit containing protodioscin was investigated in both normal and castrated rats to determine aphrodisiac effects. Rats were divided into five groups of eight animals each. The treatments were distilled water (normal and castrated animals), testosterone (normal and castrated animals, 10.0 mg/kg bw, subcutaneously, bi-weekly) and extract (castrated animals, 5.0 mg/kg bw, orally once daily for 8 weeks). Decreases in body weight and prostate weight were observed among the castrated groups of rats as compared to the normal rats. There was an overall reduction in the sexual behaviour parameters in the castrated rats as reflected by a decrease in mount and intromission frequencies and an increase in mount, intromission and ejaculation latencies as well as post-ejaculatory interval. Compared to the castrated control animals, treatment of castrated rats with either testosterone or the extract led to a mild to moderate improvement of the sexual behaviour parameters as demonstrated by an increase in mount frequency and intromission frequency and a decrease in mount latency, intromission latency and post-ejaculatory interval (*29*).

Formation of uroliths (urinary calculi)

An ethanol extract of the fruit exhibited a dose-dependent protection against uroliths induced by glass bead implantation in albino rats. The extract provided significant protection against deposition of calculogenic material around the glass bead. It also protected against leukocytosis and elevation of serum urea levels (*32*).

Toxicity

The plant, when used as fodder, has been reported to cause photosensitivity in livestock and is responsible for the diseases *geeldikkop* (in South Africa) and "bighead" (in Australia and the USA) in sheep. The condition is characterized by oedema of the head, fever and jaundice, leading to death (*6*). Two beta-carboline indoleamines (harman and norharman), present in trace amounts in the seeds of *Tribulus terrestris* have been im-

plicated in causing central nervous system effects in sheep that have grazed on *Tribulus* over a period of months. Harman and norharman appear to accumulate in tryptamine-associated neurons of the central nervous system and gradually interact irreversibly with a specific sequence of neuronal gene DNA. The extractable alkaloid content was 44.0 mg/kg dry matter. Synthetic harman and norharman administered subcutaneously to sheep at a dose of 54 mg/kg, caused nervous system effects similar to those caused by the fresh plant (*33*).

Behavioural, haematological, biochemical, functional and morphological studies on the acute, subchronic and chronic toxicities of protodioscin, one of the chemical constituents of the crude drug, showed no toxic manifestations under experimental conditions. No data were available from which conclusions could be drawn concerning carcinogenic and teratogenic effect (*33*).

Clinical pharmacology

Saponin-containing extracts of the fruit have been used to treat coronary heart disease (*22*). In a clinical observation study involving 406 patients and a cross-test (67 cases) the results showed that the extract was significantly more effective than the control for the treatment of angina pectoris, with a remission rate of 82.3% and 67.2% ($p < 0.05$), in the treatment and control groups, respectively. Electrocardiogram improvement was 52.7% in the treated group and 35.8% in the control group. The extract dilates the coronary artery and improves coronary circulation, and thus is more effective than the control treatment in improving the electrocardiogram of myocardial ischaemia. No adverse reactions were observed on haematological parameters or hepatic and renal functions (*22*).

A clinical study was conducted to assess the effectiveness of a specific dosage and period of administration of protodioscin on sperm quality and quantity in men with moderate idiopathic oligozoospermia. This study also evaluated the effect of protodioscin on libido, erection, ejaculation and orgasm. The results demonstrated that oral treatment with the dose of 3 × 2 tablets per day for 60 days increased sperm quantity and quality in men diagnosed with moderate idiopathic oligozoospermia and restored libido, erection, ejaculation and orgasm of sexual intercourse. These results were observed in more than 80% of the treated patients (*18*).

A double-blind study was done on 45 men with infertility due to oligoasthenoteratozoospermia. Thirty-six men were treated with 500.0 mg purified extract of the fruit containing protodioscin, orally 3 times daily for 3 months. The nine men in the control group were given a placebo for the same period of time. Spouses of eight of the men in the treated group became pregnant after treatment of their husbands, whereas no pregnan-

cies occurred in the spouses of the men in the control group. An improvement in the sperm morphology, acrosome morphology and reaction seemed to account for the increased fertility after treatment. In addition, the extract was shown to increase the level of dehydroepiandrosterone and might also have contributed to the activation of cell membrane receptors and the production of weak androgens (*16*).

An interesting correlation of dehydroepiandrosterone sulfate level with the incidence of low sex drive and higher occurrence of impotence was discovered in studies of patients diagnosed with diabetes mellitus. A clinical trial involving 30 men with erectile dysfunction, but not diabetes, 30 with neither erectile dysfunction nor diabetes and 15 men with both diabetes and erectile dysfunction was performed to assess the relationship between dehydroepiandrosterone sulfate and erectile dysfunction. The men were given an extract of the fruit at a dose of 3 × 250 mg per day for 3 weeks. The dehydroepiandrosterone sulfate levels, as well as other blood and liver parameters were evaluated. The results of the study showed a significant increase of dehydroepiandrosterone sulfate levels in subjects with and without diabetes after treatment, and a significant increase in the frequency of successful intercourse of 60% in subjects with or without diabetes and with or without erectile dysfunction (*17*).

Adverse reactions

No information was found.

Contraindications

Fructus Tribuli is contraindicated in cases of hypersensitivity or allergy to the plant.

Warnings

Due to the possibility of phototoxic reactions, patients using Fructus Tribuli should avoid excessive exposure to sunlight and use a sunscreen with a high sun protection factor (> 30) while taking the crude drug.

Precautions

Carcinogenesis, mutagenesis, impairment of fertility

No information was found.

Pregnancy: teratogenic effects

Due to the lack of safety data, use of the crude drug during pregnancy is not recommended.

Pregnancy: non-teratogenic effects
Due to the lack of safety data, use of the crude drug during pregnancy is not recommended.

Nursing mothers
Due to the lack of safety data, use of the crude drug during breastfeeding is not recommended.

Paediatric use
Due to the lack of safety data, use of the crude drug in children under the age of 12 years is not recommended.

Other precautions
No information was found.

Dosage forms
Crude drug and extracts.

Posology
(Unless otherwise indicated)
Oral daily dosage: 3–6 g of the powdered crude drug as a decoction (*4*); 6–9 g in divided doses daily as a decoction (*1*).

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Flos Trifolii

Definition
Flos Trifolii consists of the dried inflorescences of *Trifolium pratense* L. (Fabaceae) (*1*).

Synonyms
No information was found.

Selected vernacular names
Aasristik, aka kurooba, aka tsumekusa, akerklee, basim ahmar, beebread, broad-leaved clover, cow clover, creeping clover, hong san ye cao, hong hua san ye cao, hong che zhou cao, klever krasnyi, klever lugovoi, meadow clover, peavine clover, puna-apila, purple clover, red clover, red-klover, redo kurooba, ribah, rode klaver, rodklover, rödklöver, Rot-od-kopklee, Rothe Kleeblumen, Rother klee, Rother Wiesen-Klee, Rotklee, rod-klee, trébol, trébol common, trébol rojo, trébol violeta, tréfle common, tréfle des prés, tréfle rouge, tréfle violet, trefoil, trevor, trevo-dos-prados, trevo-violeto, trifoglio pratense, trifoglio violetto, wild red clover, Wiesen-Klee, wiesenklee (*2*–*8*).

Geographical distribution
Native to Europe. Found worldwide (*7*).

Description
A low-growing, common, perennial herb with ascending slender hairy stems bearing trifoliate leaves with broad, bristle-pointed stipules, the leaflets varying from ovate to obovate in outline, frequently notched at the apex, and showing a pale spot on their upper surface. The small butterfly-shaped flowers are borne in ovoid heads with long or short peduncles; their colour varies from magenta to whitish (*7*).

Plant material of interest: dried inflorescences

General appearance

Inflorescences are ovoid with a rounded summit, mostly from 12–34 mm in length and width, usually on a very short stalk, shrivelled, purplish, and more or less brown from drying, consisting of many papilionaceous flowers, crowded together and clothed at the base with broad, pointed, pale green ciliate stipules with darker veins. The flowers, which may or may not be accompanied by diminutive trifoliate leaves, are up to 15 mm in length and have the following: five green, hairy, subulate calyx teeth, one longer than the other four; petals united into a more or less campanulate tube, somewhat recurved, and colourless with pinkish purple veins; diadelphous stamens; slender style (*1*).

Organoleptic properties

Odour: faintly aromatic, somewhat tea-like; taste: sweetish, then slightly bitter (*1*).

Microscopic characteristics

Epidermis of calyx composed of polygonal cells with faintly striated cuticle and occasional anomocytic stomata on the outer epidermis only; abundant, uniseriate, covering trichomes with two small, thin-walled basal cells and a thick-walled tapering end cell, up to 1 mm in length with a warty cuticle. Glandular trichomes are also present, particularly on the lower epidermis, each with a one- or two-celled stalk and a large, cylindrical head composed of several cells arranged in two rows. Epidermal cells of the corolla, papillose at the tip, are elongated with slightly wavy walls and a strongly striated cuticle; vascular strands of corolla and calyx are surrounded by a crystal sheath containing prismatic crystals of calcium oxalate. The following are also present: fibrous layer of anthers; subspherical pollen grains, 20–48 mm in diameter with smooth exine, three distinct pores, and three furrows; upper epidermal cells of leaflets with sinuous and slightly beaded anticlinal walls; lower epidermis with sinuous to wavy walls; anomocytic stomata on both surfaces, but more frequent on the lower surface; abundant covering trichomes on both surfaces and on the margins; and fibrovascular strands surrounded by a crystal sheath containing prismatic crystals of calcium oxalate (*1*).

Powdered plant material

A pinkish-grey powder with a faint, fragrant odour and a slightly bitter taste. Fragments of corolla with slightly wavy walls and a striated cuticle; fragments of calyx with rectangular cells and a faintly striated cuticle; abundant uniseriate, warty-walled covering trichomes with part of the

end cell frequently broken off; glandular trichomes less frequent; subspherical pollen grains, scattered or associated with fragments of fibrous layer of anthers; abundant strands of vascular tissue with associated crystal sheath containing prismatic crystals of calcium oxalate; occasional portions of green leaf with wavy walled epidermis and anomocytic stomata (*3*).

General identity tests

Macroscopic and microscopic examination, as well as thin-layer and high-performance liquid chromatography (*1*, *3*).

Purity tests

Microbiological

Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*9*).

The United States pharmacopeia requires the absence of *Salmonella* species and *Escherichia coli*, with total aerobic microbial count not exceeding 10^6 colony-forming units (cfu) per g, the total combined moulds and yeast count should not exceed 10^4 cfu per g, and the enterobacterial count should not be more than 1000 cfu per g (*1*).

Foreign organic matter

Not more than 2% (*1*).

Total ash

Not more than 10% (*1*).

Acid-insoluble ash

Not more than 2% (*1*).

Water-soluble extractive

Not less than 15% (*1*).

Loss on drying

Not more than 12% (*1*).

Pesticide residues

The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*10*). For other pesticides, see the *European pharmacopoeia* (*10*) and the WHO guidelines for assessing quality of herbal medi-

cines with reference to contaminants and residues (*9*) and pesticide residues (*11*).

Heavy metals

For maximum limits and analysis of heavy metals, consult the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*9*). The *United States pharmacopeia* stipulates total heavy metals not more than 10 µg/g (*1*).

Radioactive residues

Where applicable, consult the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*9*).

Other purity tests

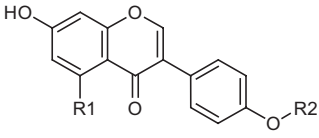
Chemical tests to be established in accordance with national requirements.

Chemical assays

Not less than 0.5% isoflavones, calculated on the dried basis as the sum of daidzein, genistein, formononetin and biochanin A (*1*).

Major chemical constituents

Rich in isoflavonoids. The major active estrogenic isoflavonoids are biochanin A, daidzein, formononetin and genistein (*5*, *13*, *14*, *17*). The structures of these isoflavonoids are presented below.



Daidzein	R1 = R2 = H
Formononetin	R1 = H R2 = CH_3
Genistein	R1 = OH R2 = H
Biochanin A	R1 = OH R2 = CH_3

Medicinal uses

Uses supported by clinical data

None.

Uses described in pharmacopoeias and well established documents

Although numerous clinical trials have assessed the safety and efficacy of red clover extracts for the treatment of menopausal symptoms, hyperlipidaemia, osteoporosis and prostate cancer (*15–19*), the data are as yet insufficient to support any of these indications. Further data from well-controlled clinical trials with sufficient numbers of subjects are needed before any therapeutic indications can be made.

Uses described in traditional medicine

Topical treatment of dermatological disorders such as psoriasis and eczema, as well as orally for the treatment of asthma and cough (*5*).

Pharmacology

Experimental pharmacology

Much of the experimental pharmacology for Flos Trifolii is based on information from pure compounds isolated from the crude drug, or from extracts of the aerial parts of the plant. Therefore, how these data apply to the crude drug needs to be further investigated.

Anti-inflammatory activity

Genistein has been found to provide protection from oxidative damage induced by ultraviolet (UV) radiation both in vitro and following dietary administration. One in vivo study assessed the potential of a number of isoflavones isolated from the crude drug, as well as some metabolically related compounds, to offer protection against UV irradiation. The results were assessed in hairless mice after topical application of the extract or isoflavones in combination with UV exposure. Daidzein, biochanin A and formononetin were not active, while 20 µM lotions of genistein and the metabolites equol, isoequol and dehydroequol reduced oedema and inflammation, as well as suppressing hypersensitivity induced by moderate doses of artificial UV radiation. The protective effect of equol was concentration-dependent and 5 µM equol markedly reduced UV-induced inflammation (*20*).

Inhibition of cell proliferation

Isoflavones inhibit the growth of some types of tumour cells, including prostate adenocarcinoma. In the prostate cancer cell line, LNCaP, and xenografts, the mechanism of the antiproliferative effects of biochanin A was determined (*18*). LNCaP cells were treated with varying concentrations of biochanin A to evaluate viability, DNA synthesis and DNA fragmentation by terminal deoxynucleotidyltransferase dUTP nick end labelling (TUNEL) analysis. Regulation of gene expression was determined using Western immunoblotting and cDNA microarrays. Antiproliferative effects were evaluated by using athymic mice with LNCaP flank tumours. Biochanin A induced a dose-dependent inhibition of LNCaP cell proliferation and tritiated thymidine incorporation that correlated with increased DNA fragmentation, indicative of apoptosis. Western blot analyses of cell cycle regulatory proteins revealed that biochanin A significantly decreased expression of cyclin B and p21, whereas flow cytometry showed that cells were accumulating in the G(0)/G(1) phase. cDNA microarray

analyses identified 29 downregulated genes with six reduced below assay detection limits. Eleven genes were upregulated, including nine that were undetectable in controls. In mice with LNCaP xenografts, biochanin A significantly reduced tumour size and incidence (*18*). In a similar study, intragastric administration of an extract of the flowers to aromatase knockout mice reduced the enlargement of a non-malignant prostate (*21*). A study examined the effect of dietary isoflavones on prostate growth in intact male mice treated with an unidentified extract of red clover. The results demonstrated that prostate, but not testis, size was significantly reduced over 28 days of being fed a diet supplemented with red clover isoflavone. Histological examination revealed an increase in apoptotic cells, rather than a reduction in proliferative activity in the epithelium. These findings support the hypothesis that red clover isoflavones in the diet can induce apoptosis and lead to a reduction in prostate size (*22*).

Estrogenic effects

The estrogenic effect of a standardized ethanol extract, with an average isoflavone content of approximately 9% (dry weight), was assessed in vitro in a yeast two-plasmid system expressing estrogen receptor alpha and estrogen receptor beta. The extract had an estrogenic activity that corresponded to a transactivational capacity of approximately 18 µg of 17-β-estradiol/g extract for estrogen receptor alpha and approximately 78 µg 17-β-estradiol/g extract for estrogen receptor beta. The difference is due to the higher binding affinity of the isoflavone constituents to estrogen receptor beta than that observed for estrogen receptor alpha (*23*).

A methanol extract (15% total isoflavones) of red clover (*Trifolium pratense* L.) showed significant competitive binding to estrogen receptor alpha and estrogen receptor beta in vitro ($p < 0.01$). In Ishikawa (endometrial cancer) cells, the extract also exhibited estrogenic activity as indicated by induction of alkaline phosphatase activity and upregulation of progesterone receptor mRNA. In S30 breast cancer cells, presenelin-2, another estrogen-inducible gene, was upregulated in the presence of red clover. Bioassay-guided isolation utilizing estrogen receptor competitive binding as a monitor, and screening using ultrafiltration liquid chromatography–mass spectrometry revealed that genistein was the most active component of red clover, and the most effective of four red clover isoflavones tested in the above in vitro assays (*24*).

An extract of the crude drug (15% total isoflavones) bound to the alpha and beta estrogen receptors with a median inhibitory concentration of 18 and 2 µg/ml, respectively. The extract activated the estrogen response element in Ishikawa cells and induced luciferase expression in MCF-7 cells (*25*).

An in vivo study assessed the estrogenic effects of the crude drug in ovariectomized rats (*25*). A red clover extract, standardized to contain 15% isoflavones was administered by gavage, at a dose of 250, 500 or 750 mg/kg body weight (bw) per day, to virgin, ovariectomized 50-day-old rats, for 21 days in the presence and absence of 17-β-estradiol (50 µg/kg bw per day). The estrogenic effects assessed included an increase in uterine weight, vaginal cell cornification and branching of mammary gland ducts. The extract of the flowers produced a dose-dependent increase in uterine weight and differentiated vaginal cells at the two higher doses, but it did not stimulate cell proliferation in the mammary glands. Neither antiestrogenic nor additive estrogenic properties were observed in any of the tissues studied. These data suggest that red clover extract is weakly estrogenic in ovariectomized rats (*26*).

Thyroid effects

The effects of isoflavones on the secretion of thyroid hormones as well as on the immunoreactivity to estrogen receptor alpha in the thyroid glands of ovariectomized ewes were studied. Eight ewes were fed 3.5 kg of 100% red clover silage daily for 14 days. Blood samples were collected before and on day 14 of exposure to phytoestrogens. After 5 months, four of the ewes were re-exposed to red clover silage as described above and the other four served as controls. Ewes exposed to red clover silage had significantly higher plasma concentrations of total T(3) and free T(3) than ewes fed hay. The cross-sectional area of thyroid follicles tended to be larger in ewes fed red clover silage than in the control animals. Estrogen receptor alpha immunoreactivity was stronger in thyroid glands from ewes exposed to phytoestrogens than in ewes fed hay. Daily ingestion of 81–95 mg phytoestrogens per kg bw for 14 days stimulated secretion of thyroid hormones and tended to increase follicle size and estrogen receptor alpha immunoreactivity of thyroid glands (*27*).

Clinical pharmacology

A randomized, double-blind, placebo-controlled, cross-over trial involving 51 perimenopausal and postmenopausal women assessed the effects of an extract of the crude drug for the treatment of hot flushes (*28*). The subjects had been amenorrhoeic for at least 6 months and had at least three hot flushes per day. The women received one tablet of either placebo or the extract (containing 40 mg total isoflavones, including genistein, 4.0 mg; daidzein, 3.5 mg; biochanin, 24.5 mg; and formononetin, 8 mg). Phase one of the trial lasted for 3 months, and was followed by a 1-month washout period. The subjects were then crossed over to the other arm for a further 14 weeks. All subjects were required to maintain a diary of

symptoms based on the Greene Menopause Score list. Of the initial 51 subjects entering the trial, 43 completed the study. At 12 weeks the frequency of hot flushes had decreased in both the group receiving placebo and in the group receiving the treatment, by 18% and 20%, respectively. However, there were no statistically significant differences between groups in frequency of hot flushes or Greene Scores at any time point. No significant changes in body weight, steroid hormone binding globulin levels, blood counts, serum electrolytes, urea, creatinine or liver function were observed (*28*).

A second randomized, double-blind, placebo-controlled trial involved 37 perimenopausal and postmenopausal women and also assessed the effects of an extract of the crude drug for the treatment of hot flushes (*15*). All subjects had been amenorrhoeic for at least 6 months and had at least three hot flushes per day. The women were randomly assigned to receive either one tablet of placebo or one of two doses of an extract (containing 40 mg or 160 mg of total isoflavones; the 40-mg tablet containing genistein, 4.0 mg; daidzein, 3.5 mg; biochanin, 24.5 mg; and formononetin, 8 mg) for 12 weeks. The outcomes measured were similar to those of the previous study. During the 12-week treatment period, the frequency of hot flushes was reduced by 35% in the placebo group; by 29% in the group treated with 40 mg; and 34% in the group treated with 160 mg of the extract. No significant changes in body weight, levels of steroid hormone binding globulin, blood counts, serum electrolytes, urea, creatinine or liver function were observed (*15*).

A third randomized, placebo-controlled trial involving 30 menopausal women who had had amenorrhoea for more than 12 months, and who were also experiencing more than five hot flushes per day, assessed the effect of the same extract as described in the previous two studies for the treatment of menopausal symptoms. All subjects participated in a single-blind phase in which they received placebo tablets for 4 weeks, and then they were randomly assigned to receive either placebo or 80 mg isoflavones for a further 12 weeks. Efficacy was measured by the decrease in number of hot flushes per day and changes in the Greene Menopause Scale score. During the first 4 weeks of treatment with the placebo, the frequency of hot flushes decreased by 16%. During the subsequent double-blind phase, a further, statistically significant decrease of 44% was seen in the group treated with 80 mg of isoflavones ($p < 0.01$), whereas no further reduction occurred in women in the group treated with the placebo. The Greene Menopause Scale score decreased by 13% in the group treated with the isoflavones and remained unchanged in the women treated with the placebo (*29*).

A fourth study compared the efficacy and safety of two products derived from red clover with that of a placebo in symptomatic menopausal women. This randomized, double-blind, placebo-controlled trial involved menopausal women, aged 45–60 years, who were experiencing at least 35 hot flushes per week. The study included women who had recently become postmenopausal (mean (standard deviation), 3.3 (4.5) years since menopause) experiencing on average 8.1 hot flushes per day. Women were excluded from the study if they were vegetarians, consumed soy products more than once per week, or took medications affecting isoflavone absorption. After a 2-week placebo run-in, 252 participants were randomly assigned to receive either Promensil (82 mg of total isoflavones per day), Rimostil (57 mg of total isoflavones per day) or a placebo identical in appearance to the clover products, and followed up for 12 weeks. The primary outcome measured was the change in frequency of hot flushes as recorded by participants in their daily diaries. Secondary outcome measures included changes in quality of life and adverse events. Of 252 participants, 246 (98%) completed the 12-week protocol. The reductions in mean daily count of hot flushes at 12 weeks were similar in all three groups: Promensil, 5.1; Rimostil, 5.4; and placebo, 5.0. In comparison with the group treated with the placebo, participants who received the Promensil (41%; 95% confidence interval (CI), 29%–51%; $p = 0.03$), but not those treated with Rimostil (34%; 95% CI, 22%–46%; $p = 0.74$) found that the treatment reduced hot flushes more rapidly. Improvements in quality of life and reports of adverse events were comparable in all three groups (*19*).

Data from two clinical trials, published only as abstracts, are also available (*30*). A randomized, double-blind, placebo-controlled pilot study conducted in Peru assessed the efficacy of a red clover product in 30 postmenopausal women. The women were treated with 40 mg of the extract or a placebo for 4 months (*31*). A 75% reduction in hot flushes occurred in the treatment group; however, no data for the placebo group were reported. The second study was an uncontrolled trial (no placebo group) in 23 women with ammenorrhoea for 12 months (*32*). In patients treated with 40.0 mg of the extract, there was a 56% reduction in the frequency of hot flushes over a 2-month period. The severity of hot flushes decreased by 43% and the severity of night sweats decreased by 52%. No changes in endometrial thickness and no adverse effects were observed. Complete blood counts were all within normal limits (*32*).

The effects of a red clover-derived isoflavone extract on the Ki-67 proliferative marker of endometrial biopsies were assessed in a double-blind, randomized, controlled study involving 30 perimenopausal women. The

purpose of the study was to detect a decrease in the Ki-67 proliferative index during the late follicular phase after a 3-month course of 50.0 mg/day red clover isoflavones. The biopsies were timed as close as possible to days 7–11 of the menstrual cycle, and simultaneous measurements of trans-vaginal endometrial thickness, uterine artery Doppler, hormone profiles, lipids and bone markers were made. Of 30 women, two did not return for a second biopsy, and a third had an unsuccessful second biopsy. Four subjects were excluded from the intention-to-treat analysis because they did not have menstrual bleeding within the time frame of the study (3 subjects) or were tested on day 13 instead of between days 7 and 11 of the cycle (1 subject). There was no change in the Ki-67 proliferation index after treatment in either group. Eight women in the group given the placebo and eight in the treatment group had proliferative endometrial biopsies that were synchronized with estradiol levels at baseline and post-treatment, and analysis of these subjects revealed no detectable change in the relationship between estradiol levels and Ki-67 with treatment in either group. There was no change in fasting lipids, bone markers, uterine Doppler resistance or pulsatility index (*33*).

Effects on bone and cardiovascular system

Six trials have assessed the effects of isoflavones on total cholesterol and lipid levels (*27*, *34–38*). A randomized, single-blind, cross-over study involving 21 premenopausal women with regular menstrual cycles assessed the effect of an extract of the crude drug on plasma lipids and oxidization of low-density lipoprotein cholesterol (*34*). Subjects were treated with a placebo or 86 mg of isoflavones per day for two menstrual cycles, after which they were crossed over to the other group. Fourteen women completed the study and no differences in total cholesterol, triglycerides or oxidized low-density lipoprotein were observed.

A randomized, double-blind, placebo-controlled study involving 66 postmenopausal women with hypercholesterolaemia assessed the effects of an ascending dose of the extract, 40 or 80 mg of isoflavones, on lipid profiles (*35*). Study participants were asked to follow a low isoflavone diet for 3 months during the study. Treatment did not affect total cholesterol, low-density lipoprotein, high-density lipoprotein cholesterol or plasma triglycerides at any dose.

A double-blind placebo-controlled trial compared the effects of two doses of isoflavones, 40 and 80 mg, with the effects of a placebo on arterial compliance and plasma lipids in menopausal women. After a 3–4-week run-in period and a 5-week placebo phase, 26 women were randomly allocated to receive either 40 mg of the extract or the placebo for an additional 5 weeks, and then the dose was increased to 80 mg. Sixteen

women completed the trial. The results demonstrated a significant improvement in arterial compliance at both the 40- and 80-mg doses as compared with placebo ($p < 0.05$). No significant differences were seen between the two doses (*30*, *36*).

A double-blind study to evaluate the effects of varying doses of isoflavones on lipid and bone metabolism in postmenopausal women was performed. An extract of red clover, containing genistein, daidzein, formononetin and biochanin was administered to 46 postmenopausal women after a single-blind placebo phase; this was followed by a single-blind wash-out phase. Patients were randomly assigned to receive 28.5 mg, 57 mg or 85.5 mg of isoflavones daily for 6 months. At 6 months, the serum high-density lipoprotein cholesterol was found to have risen significantly by 15.7–28.6% with the different doses ($p = 0.007$, $p = 0.002$ and $p = 0.027$ at doses of 28.5 mg, 57 mg and 85.5 mg, respectively), although the magnitude of the response was independent of the dose used. The serum apo-lipoprotein B fell significantly by 11.5–17.0% with the different doses ($p = 0.005$, $p = 0.043$, $p = 0.007$, respectively) and the magnitude of the response was independent of the dose used. The bone mineral density of the proximal radius and ulna rose significantly by 4.1% over 6 months in the women who received 57 mg/day ($p = 0.002$) and by 3.0% in the women who received 85.5 mg/day ($p = 0.023$) of isoflavones. The response to treatment with 28.5 mg/day of isoflavones was not significant. These results show that the administration of an isoflavone combination extracted from red clover was associated with a significant increase in high-density lipoprotein cholesterol, a significant fall in apolipoprotein B, and a small but significant increase in the predominantly cortical bone of the proximal radius and ulna after 6 months of treatment (*37*).

A double-blind, placebo-controlled study, involving 107 premenopausal, perimenopausal and postmenopausal women assessed the effects of an extract of the crude drug (corresponding to 40 mg of isoflavones per day) for 1 year on bone mineral density and content (*39*). The outcomes measured were changes in the lumbar spine and total hip bone mineral content and density. After 1 year, the bone mineral content and density of the lumbar spine (measured by dual energy X-ray absorptiometry (DEXA) scan) had decreased significantly ($p < 0.01$) in both groups. However, the decrease in the bone mineral content and density of the spine of premenopausal and perimenopausal women treated with the extract was significantly lower than that in women who received the placebo ($p < 0.02$ and $p < 0.01$, respectively). No differences were observed in the postmenopausal women. No differences in bone mineral density of the

hip or significant changes in markers of bone turnover were seen in any of the groups (*39*).

A 12-week randomized, double-blind, placebo-controlled trial was conducted involving 252 menopausal women aged 45–60 years. The women, who were experiencing > 35 hot flushes per week, were randomly assigned to receive either Promensil (82 mg total isoflavones), Rimostil (57.2 mg total isoflavones) or a placebo. The primary outcome measures were mean absolute changes for high-density lipoprotein cholesterol, serum osteocalcin and urinary N-telopeptide. The secondary outcome measures were mean changes of total cholesterol, low-density lipoprotein cholesterol, the ratio of high-density lipoprotein cholesterol to low-density lipoprotein cholesterol, and triglycerides. Ninety-eight per cent of the participants completed the 12-week protocol. Women who took Rimostil or Promensil had greater mean increases in high-density lipoprotein cholesterol than those who took placebo; however, this change was small (< 2 mg/dl) and was not statistically significant. There was a significant decrease in triglyceride levels among women who took Rimostil (14.4 mg/dl) or Promensil (10.9 mg/dl) compared to those who took the placebo. The decrease was seen primarily among women with elevated baseline triglyceride levels. There were no differences in mean changes of total cholesterol, low-density lipoprotein cholesterol, or the ratio of high-density lipoprotein cholesterol to low-density lipoprotein cholesterol among treatment groups. There were no statistically significant differences between treatment groups for bone turnover markers. It was concluded that compared with placebo, extracts containing isoflavones decrease levels of triglycerides in symptomatic menopausal women; however, this effect was small (*38*).

Treatment of prostate cancer

A nonrandomized, non-blinded trial with historically matched controls from archival tissue assessed the effects of acute exposure to a dietary supplement of isoflavones in men with clinically significant prostate cancer before radical prostatectomy. Thirty-eight patients were recruited to the study upon diagnosis of prostate cancer. Before surgery, 20 of the men consumed 160 mg/day of red clover-derived dietary isoflavones, containing a mixture of genistein, daidzein, formononetin and biochanin A. Serum prostate-specific antigen, testosterone and biochemical factors were measured, and clinical and pathological parameters were recorded. The incidence of apoptosis in prostate tumour cells from radical prostatectomy specimens was compared between 18 treated and 18 untreated control tissues. There were no significant differences between pretreatment and post-treatment serum prostate-specific antigen, Gleason score, serum

testosterone or biochemical factors in the treated patients ($p > 0.05$). Apoptosis in radical prostatectomy specimens from treated patients was significantly higher than in control subjects ($p = 0.0018$), specifically in regions of low to moderate-grade cancer (Gleason grade 1–3). No adverse events related to the treatment were reported (*18*).

Pharmacokinetics and pharmacodynamics

The pharmacokinetics of red clover isoflavones were investigated after long-term administration as a once-daily dietary supplement. Fourteen subjects who had been consuming a low-isoflavone containing diet for 2 weeks were given an oral dose of two isoflavone tablets (approximately 80 mg of total isoflavones) daily for 2 weeks. Before the study day the participants fasted overnight and were then administered their last dose the next morning. Plasma samples were collected for a 48-hour period after the last dose. Plasma isoflavones were assayed by high-performance liquid chromatography. The results demonstrate that trough plasma levels were significantly higher for daidzein and genistein after long-term dosing than levels recorded prior to the commencement of the study and plasma levels of isoflavones after long-term dosing were in the range previously reported in populations that consume an isoflavone-rich diet. The plasma half-lives observed after long-term administration were consistent with once-daily administration. Isoflavones have pharmacokinetic characteristics that suggest that once-daily administration is adequate when they are administered long-term (*40*).

The absorption of isoflavones varies substantially between individuals. A single-blind, randomized, placebo-controlled, cross-over trial involving 14 subjects investigated whether isoflavone absorption differs depending on whether the isoflavones originated from soy beans or from red clover. Soy bean isoflavone glycosides and red clover isoflavone aglycones were incorporated into a breakfast cereal and eaten daily for 2 weeks each, separated by a 2-week control or washout period. The excretion of isoflavones in urine was measured over a 24-hour period; approximately 25% of each isoflavone was recovered in urine, suggesting that similar amounts were absorbed irrespective of their glycoside/aglycone nature or the differing compositions of their sources (daidzein and genistein in soy beans and formononetin and biochanin in red clover). Although interindividual variability was high, there was less intraindividual variability; the amounts excreted when subjects consumed the two sources of isoflavone were correlated ($r = 0.69$; $p = 0.007$) (*41*).

Adverse reactions

Ingestion of large amounts of clover in animal feed has been associated with a number of adverse effects in sheep in Australia. A publication on "clover disease" described symptoms of infertility, abnormal lactation, dystocia and prolapsed uterus, all of which were hypothetically attributed to the estrogenic effects of isoflavones (*42*). None of the controlled, clinical trials has reported adverse effects at doses up to 160 mg of isoflavones per day. *Trifolium pratense* does not contain coumarins, and therefore the concerns about blood coagulation are unfounded (*43*, *44*).

Contraindications

Flos Trifolii is contraindicated in cases of hypersensitivity or allergy to the crude drug. It is also contraindicated during pregnancy, breastfeeding and for children under the age of 12 years, and in cases of hormone-associated diseases, due to the potential hormonal effects.

Warnings

Due to the potential estrogenic effects of the crude drug, patients with hormone-related disorders, estrogen-dependent cancers or a familial history of estrogen-dependent cancers should contact a health care provider before use.

Precautions

Carcinogenesis, mutagenesis, impairment of fertility

No information was found.

Drug interactions

There are conflicting data concerning an interaction of the crude drug with tamoxifen and other antiestrogenic drugs. Some studies suggest that specific isoflavones may enhance the ability of tamoxifen to inhibit the growth of estrogen-receptor-positive breast cancer cells (*45–47*). In rodent models, genistein has been shown to inhibit the efficacy of tamoxifen on the growth of estrogen-receptor-positive breast cancer cells implanted in ovariectomized mice, while other research shows that specific isoflavones may be additive and work in a synergistic manner to prevent the development of chemically induced tumours and the growth of existing tumours (*48–50*). Therefore, the use of the crude drug or its preparations is not recommended in those being treated with tamoxifen and other antiestrogenic drugs until further research has been done.

Pregnancy: non-teratogenic effects
See Contraindications.

Nursing mothers
See Contraindications.

Paediatric use
See Contraindications.

Dosage forms
Crude drug and tablets.

Posology
(Unless otherwise indicated)
Oral dose: extracts of crude drug: 240–480 mg corresponding to 40–80 mg/day of isoflavones (*15*, *17*, *28*, *29*).

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Ramulus cum Uncis Uncariae

Definition
Ramulus cum Uncis Uncariae consists of the dried hook-bearing stem branch of *Uncaria rhynchophylla* (Miq.) Jacks, *U. macrophylla* Wall., *U. hirsuta* Havil., *U. sinensis* (Oliv.) Havil. or *U. sessilifructus* Roxb. (Rubiaceae) (*1*–*3*).

Synonyms
Uncaria rhynchophylla (Miq.) Jacks, *Nauclea rhynchophylla* Miq., *Ourouparia rhynchophylla* Matsum (*4*).

Selected vernacular names
Choto-ko, chotoko, cho-to-kou, gout eng, kagikazura (*1*, *4*, *5*).

Geographical distribution
Indigenous to China and Japan (*6*, *7*).

Description
A deciduous twining shrub up to 10 m long. Branches brownish, glabrous, occasionally bearing compressed, hooked thorns. Young stems slender, square to slightly angular, glabrous. Stipules of the plagiotropic shoot 6–10 mm long, those of the orthotropic shoot considerably larger, up to 30 mm long, inside glabrous with glandular hairs at the base, outside glabrous, margins entire, narrowly triangular, deeply bifid for over two thirds of the length, lobes narrowly triangular to triangular-lanceolate. Leaves opposite, ovate to ovate-oblong or elliptic to elliptic-oblong, 5–12 × 3–7 cm, membranous, glabrous on either side; apex acute to cuspidate; base cuneate to truncate; lateral nerves 4–8 pairs, axils with sparsely hairy domatia, tertiary nerves curved, impressed, ultimate venation reticulate. Inflorescence an axillary or terminal peduncled solitary head. Flowers yellow, subsessile on the receptacle. Hypanthium 1–2 mm, densely hairy. Calyx 1 mm, pubescent; lobes oblong to slightly triangular, 0.5–1 mm, sparsely pubescent. Corolla tube 6–8 (6–10 mm), outside

glabrous or with a few scattered hairs; lobes oblong, 1.5–2.5 mm, outside glabrous or slightly farinose-pubescent, margins sometimes ciliate. Stamens 5. Ovary 2-celled. Fruiting head 14–18 mm in diameter, fruit a dry capsule. The plant entwines other trees with its hooked thorns (*6*, *8*, *9*).

Plant material of interest: dried hook-bearing stem branch

General appearance

Cylindrical or subsquare, 1–4 cm long, 2–5 mm in diameter. Externally reddish-brown to dark brown or yellow-brown with fine longitudinal striations, glabrous, or yellowish-green to greyish-brown, sometimes with white dotted lenticels, covered with yellowish-brown pubescence. Transverse section: square to elliptical. Most nodes of stem branches with two opposite downward curved hooks/prickles (sterile peduncles), some with a hook only on one side and with raised scars on the other side; hooks relatively flattened or rounded, apex acute, base relatively broad; dotted scars of fallen petiole and ring-shaped stipule scars visible on the branch at the hook base; transverse section oblong to elliptical. Texture hard and tenacious, fracture yellowish-brown, bark fibrous, pith yellowish-white to light brown or hollowed (*1*, *2*).

Organoleptic properties

Odour: none; taste: slight (*1*, *2*).

Microscopic characteristics

Transverse section of the prickle (hook) reveals vascular bundles in the cortex, unevenly distributed and arranged in a ring. Parenchyma cells in the secondary cortex containing crystals of calcium oxalate (*2*, *3*).

Powdered plant material

To be established in accordance with national requirements.

General identity tests

Macroscopic examination (*1–3*), microchemical test (*1–3*), and high-performance liquid chromatography for the presence of characteristic oxindole alkaloids (*2*, *10–13*).

Purity tests

Microbiology

Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*14*).

Chemical
To be established in accordance with national requirements.

Foreign organic matter
Not more than 2% (*3*).

Total ash
Not more than 4% (*2*, *3*).

Acid-insoluble ash
To be established in accordance with national requirements.

Water-soluble extractive
To be established in accordance with national requirements.

Alcohol-soluble extractive
Not more than 6% (*1*).

Loss on drying
Not more than 12% (*2*).

Pesticide residues
The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*15*). For other pesticides see the *European pharmacopoeia* (*15*) and the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*14*) and pesticide residues (*16*).

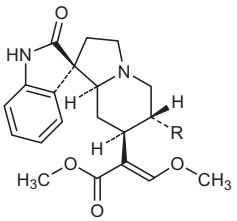
Heavy metals
For maximum limits and analysis of heavy metals, consult the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*14*).

Radioactive residues
Where applicable, consult the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*14*).

Chemical assay
Not less than 0.03% of total rhynchophylline (rhynchophylline and hirsutine) as determined by high-performance liquid chromatography and calculated on the dried basis (*2*).

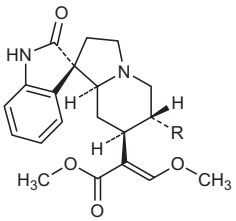
Major chemical constituents
The major constituents are indole alkaloids (1.7%) including the tetracyclic oxindoles rhynchophylline (0.02%), isorhynchophylline (0.05%),

corynoxeine (0.006%) and isocorynoxeine (0.001%). Other notable alkaloids are hirsuteine (0.001%) and hirsutine (0.001%). Besides ursolic acid, two other triterpenes, the bioactive uncarinic acids A and B as well as the flavonoid, hyperin, have been reported (*4*, *5*, *17*). Structures of major tetracyclic indole and oxindole alkaloids and the triterpenes uncarinic acids A and B are presented below.



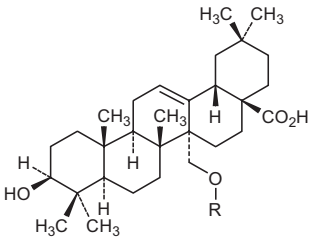
Corynoxeine R = $CH{=}CH_2$

Rhynchophylline R = $CH_2{-}CH_3$



Isocorynoxeine R = $CH{=}CH_2$

Isorhynchophylline R = $CH_2{-}CH_3$

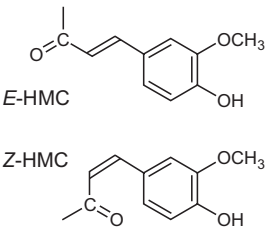


Uncarinic acid A

R = *E*-HMC

Uncarinic acid B

R = *Z*-HMC



Medicinal uses

Uses supported by clinical data

None.

Uses described in pharmacopoeias and well established documents

Used orally to treat eclampsia, headache, dizziness, high fevers and hypertension (*1*, *6*). Considering the serious nature of these diseases, the usefulness of the crude drug needs to be confirmed by controlled clinical studies.

Uses described in traditional medicine

Used as a carminative, diuretic, and a muscle relaxant. Also used to treat cardiovascular disease, colic, convulsions, stroke and vertigo (*5*, *6*).

Pharmacology

Experimental pharmacology

Much of the pharmacological work has been performed with pure compounds isolated from the crude drug. These studies used high concentra-

tions of the pure compounds and thus the applicability of these data to the crude drug and its preparations needs to be further investigated.

Antiarrhythmic activity

The effect of hirsutine, an indole alkaloid isolated from the crude drug, on membrane potentials of rabbit sino-atrial node and guinea-pig right ventricle and left atrium was investigated. Hirsutine at concentrations of 0.1–30.0 μM decreased the maximum rate of rise and prolonged action potential duration in sino-atrial node in vitro, as well as in atrial and ventricular preparations (*18*). Hirsutine, in concentrations of 1.0–3.0 μM produced a dose-dependent relaxation of the isolated rat aorta precontracted with either norepinephrine or potassium chloride solution, suggesting that the compound dilated blood vessels through the inhibition of voltage-dependent calcium channels (*19*). The compound, at a concentration of 30 μM, also decreased intracellular calcium concentrations in isolated vascular smooth muscle cells through the inhibition of norepinephrine-induced calcium influx (*20*).

Anticonvulsant activity

Intragastric administration of a methanol extract of the crude drug to rats at a dose of 1.0 g/kg body weight (bw) inhibited kainic acid-induced epileptic seizures and reduced the levels of free radicals, as measured by lipid peroxidation in the brain (*21*).

Intraperitoneal administration of an ethyl acetate or methanol extract of the crude drug to mice at a dose of 70.0 mg/kg bw inhibited pentetrazole-induced convulsions (*22*). Hyperin, a chemical constituent isolated from the extract, decreased the elevated activities of γ-aminobutyric acid-T, xanthine oxidase and lipid peroxide induced by pentetrazole in mouse brain tissue in vitro at a concentration of 25.0 mg/ml (*23*).

Intragastric administration of an aqueous extract of the crude drug to mice, at a dose of 1.0–3.0 g/kg bw, inhibited glutamate-induced convulsions, but had no effect on convulsions induced by picrotoxin, strychnine or electroshock (*24*). The active constituents of the crude drug were isolated and identified as indole alkaloids, geissoschizine methyl ether and hirsutine. Intragastric administration of the alkaloids to mice at a dose of 100.0 mg/kg bw also inhibited glutamate-induced convulsions in a dose-dependent manner (*24*).

An aqueous extract of the crude drug at a concentration of 0.1 mg/ml prevented glutamate-induced neuronal death in cultured rat cerebellar granule cells through the inhibition of calcium influx into the cells (*25*). In cultured mouse cerebral neurons, glucose oxidase-induced toxicity was reduced after treatment of the cells with 100 μg/ml of the crude drug (*26*).

In cultured hippocampal neurons from neonatal mice, hydrogen peroxide-induced neuronal damage was significantly reduced after treatment of the cells with 80 µg/ml of the crude drug (*27*). A methanol extract of the crude drug protected against N-methyl-D-aspartate-induced excitotoxicity in cultured rat hippocampus slices at a concentration of 100 µg/ml (*28*).

Antihypertensive activity

An alkaloid-containing extract of the crude drug reduced blood pressure in rats when administered by gavage at a dose of 50.0 mg/kg bw for 20 days, or when administered by intravenous injection to cats at a dose of 20.0 mg/kg bw for 20 days (*29*). A 50% methanol extract of the crude drug inhibited the activity of angiotensin-converting enzyme isolated from pig kidney at a concentration of 200.0 µg/ml in vitro (*30*).

An infusion of the crude drug inhibited norepinephrine-induced contractions of isolated rat aorta at a concentration of 0.12 mg/ml (*31*). Endothelium-dependent relaxation induced by the extract of the crude drug was inhibited by *N*-monomethyl-L-arginine, but not indometacin or atropine, and was decreased when the endothelium was not present. The authors concluded that the extract relaxes the precontracted rat aorta through an endothelium-dependent mechanism involving nitric oxide (*31*).

Anti-inflammatory activity

A decoction of the crude drug suppressed carrageenan-induced footpad oedema in rats when administered by subcutaneous injection at a dose of 10.0 ml/kg bw (*32*). An infusion of the crude drug inhibited the activity of prostaglandin synthetase in rabbit microsomes at a concentration of 750.0 µg/ml (*33*).

Antimicrobial activity

An ethanol extract of the crude drug was not active against *Bacillus subtilis*, *Candida albicans*, *Escherichia coli* or *Streptococcus pneumoniae* in vitro at concentrations up to 500.0 mg/disc (*34*).

Antioxidant activity

Intraperitoneal administration of an ethanol extract of the crude drug at a dose of 1.0 g/kg bw to rats inhibited an increase in lipid peroxidation in the ipsilateral cortex induced by the injection of ferric chloride into the lateral cortex (*35*). The extract also induced an increase in the activity of superoxide dismutase in the mitochondrial fraction of the ipsilateral cortex (*35*). A methanol extract of the crude drug inhibited kainic acid-induced lipid peroxidation in rat brain tissues in vitro (*21*). An aqueous extract of the crude drug, at a concentration of 50.0 µg/ml, had strong

scavenging activity against superoxide anion radicals in vitro, as measured by electron spin resonance spin-trapping techniques (*36*).

Depressant effects on the central nervous system

Intragastric administration of an aqueous extract of the crude drug (2.0 g/kg bw), or oxindole alkaloids isolated from the crude drug (10–100 mg/kg bw) to mice, significantly prolonged thiopental-induced sleep time ($p < 0.05$) (*13*, *37*). Intragastric administration of an aqueous extract of the crude drug to mice at a dose of 2.0 g/kg bw significantly depressed locomotor activity ($p < 0.05$) (*37*). Intragastric administration of three indole alkaloids isolated from the crude drug, corynoxine (30.0 mg/kg bw), corynoxine B (100.0 mg/kg bw) or isorhynchophylline (100.0 mg/kg bw) also significantly decreased locomotor activity ($p < 0.05$) (*38*).

The suppressant effects on the central nervous system were confirmed by a study in mice of extracts prepared from mixtures of the herb with and without the crude drug. Preparations containing the crude drug prolonged thiopental-induced sleep, but the extract prepared from the mixture of herb without the crude drug was devoid of this activity (*39*). Intraperitoneal administration of a 90% ethanol extract of the crude drug reduced spontaneous motor activity in mice (*34*).

Effects on neurotransmitters

An 80% ethanol extract of the crude drug inhibited serotonin reuptake by 97% at a concentration of 10.0 µg/ml in rat brain stem neurons (*40*). Uncarinic acids A and B, isolated from the crude drug inhibited the activity of phosphatidylinositol-specific phospholipase C, a key enzyme involved in the signal transduction of growth factors, neurotransmitters and hormones, with a median inhibitory concentration of 35.66 and 44.55 µM, respectively (*41*). Hirsutine, at a concentration of 300 nM to 10.0 µM inhibited dopamine release induced by nicotine in rat pheochromocytoma PC12 cells (*42*). In a study in rats, dopamine release induced by potassium chloride was also inhibited by hirsutine at a concentration of 10.0 µM (*41*). Hirsutine also had antagonistic effects on the opioid receptors and, at a concentration of 10.0 µM, reversed the inhibitory effect of morphine on twitch contraction in isolated guinea-pig ileum (*43*).

Toxicology

Intraperitoneal administration of a 90% ethanol extract of the crude drug to mice had a median lethal dose of 1.0 g/kg bw (*34*).

Clinical pharmacology

No information was found.

Adverse reactions
No information was found.

Contraindications
Allergy to the plant material.

Warnings
No information was found.

Precautions
General
No information was found.

Drug interactions
No information was found.

Drug and laboratory test interactions
No information was found.

Carcinogenesis, mutagenesis, impairment of fertility
An aqueous extract of the crude drug was not mutagenic in the Ames test at a concentration of 40.0–50.0 mg/plate in *Salmonella typhimurium* strains TA98 and TA100 (*44*, *45*). The extract was also not mutagenic when administered by intraperitoneal injection to mice at a dose of 4.0 mg/kg bw, equal to 10–40 times the amount used in humans (*44*). An infusion of the crude drug inhibited aflatoxin B1-induced mutagenesis in *Salmonella typhimurium* strains TA98 and TA100 at a concentration of 40.0 mg/plate (*46*). Metabolic activation was required for activity.

Pregnancy: teratogenic effects
No information was found.

Pregnancy: non-teratogenic effects
Use of the crude drug during pregnancy requires the supervision of a health care professional.

Nursing mothers
No safety data are available, thus the use of the crude drug by breastfeeding mothers is not recommended.

Paediatric use
No safety data are available, thus the use of the crude drug in children under the age of 12 years is not recommended.

Other precautions
No information was found.

Dosage forms
Crude drug (*1*). Store in a dry place (*1*).

Posology
(Unless otherwise indicated)
Oral daily dose: 3–12 g of the crude drug, added to a decoction, taken in divided doses (*1*).

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Cortex Viburni Prunifolii

Definition
Cortex Viburni Prunifolii consists of the dried root or stem, trunk or root bark of *Viburnum prunifolium* L. (Caprifoliaceae) (*1–3*).

Synonyms
Viburnum pyrifolium Poiret, *V. prunifolium* var. *globosum* Nash., *V. lentago* var. *pyrifolium* (Poiret) Chapman, *V. bushii* Ashe, *V. prunifolium* var. *bushii* (Ashe) Palmer and Steyermark (*3*).

Selected vernacular names
Acerola negra, American sloe, American snowball, Amerikanische Viburnum, Amerikanischer Schneeball, Amerikansk snebolles, baya negra, black haw, black hawthorn, Blommenbladt kvalkved, cramp bark, ploomilehine lodjapuu, sheepberry, sloe, sloe tree, stagberry, stagbush, sweet viburnum, viburno americano, Virginische Schneeball Schneebaum (*2–5*).

Geographical distribution
Native to the USA (*2–4*, *6*).

Description
A shrub or small tree up to 8 m in height. Bark is dark grey, lustrous and flaking. Leaf: deciduous, alternate, broadly elliptic to ovate-oblong, rarely lanceolate, 1.8–6.2 cm (mostly 2.3–4.0 cm) wide, 2.5–9.7 (mostly 3–6) cm long, apex rounded or acute, base obtuse to decurrent, surface membranous or chartaceous, glabrous with stellate hairs along top midrib, and margins serrulate. Petioles 10–12 (occasionally 4–18) mm long, narrowly winged or not. Inflorescence: terminal, many-flowered cyme, flat-topped, 3.5–12 cm across, bracts 5, peduncle up to 11 mm long. Flower: perfect, radially symmetric, spreading to broadly campanulate; sepals and petals 5, each whorl partially fused; corolla white, lobes 2–3 mm long; stamens 5, exerted, anthers yellow; ovary inferior, 3-carpellate with only 1 locule fer-

tile, uni-ovulate, style short with 3 sessile stigmas. Fruit**:** drupe globose, blue-black, glaucous, 6–10 mm wide, 7–16 mm long, persistent (*3*, *6*).

Plant material of interest: dried stem, trunk and root bark

General appearance

Stem and trunk bark: irregularly and transversely curved or quilled or cut into irregularly shaped oblong chips of varying sizes (1.5–15 cm in length by 0.5–1.5 cm in diameter, and up to 6 mm in thickness). The outer surface of young bark is silvery grey with raised oval lenticels. The outer bark is usually covered with lichens and mosses. The outer surface of older bark is greyish-brown to black or reddish-brown where the cork has been abraded. When cork is present, usually on older bark, it is divided into irregular oblique, longitudinal, or transverse intersecting fissures.

The inner surface is longitudinally striated, pale yellowish to reddish-brown, or pale yellow with reddish-brown blotches and streaks. The fracture is short and uneven showing greyish to blackish outer bark, greenish-brown to reddish-brown middle bark and light brown to whitish inner bark in which scattered groups of pale yellowish stone cells may be discerned with a hand lens (*3*, *6*).

Root bark: transversely curved pieces or quills 1.5–10 cm long, up to 2 cm wide and 0.5–4 mm thick. The outer surface is greyish-brown or brownish-red where the cork is missing. The youngest pieces have slight longitudinal wrinkles, older bark has small rounded or oval lenticels and is irregularly wrinkled, fissured and scaly. The inner surface is pale yellowish to reddish-brown with longitudinal striations. The fracture is weak, brittle, short and uneven. The fractured surface shows greyish-brown or dark brown cork, brownish to brownish-red middle bark, and whitish inner bark with a number of pale yellow groups of stone cells visible with a hand lens. Fractured surfaces exhibit numerous minute calcium oxalate crystals that appear as glittering points (*3*, *6*).

Organoleptic properties

Odour: characteristically pungent, valerian-like; taste: astringent and bitter (*3*, *6*).

Microscopic characteristics

Stem bark: cork of variable width and composed of usually tangentially elongated cells with suberized to lignified walls. The cork layer consists of reddish-brown polygonal cells. The primary cortex consists of slightly thickened parenchyma cells containing calcium oxalate rosettes (10–20 µm in diameter). Oil droplets are present in the bark of young plants. When

viewed in cross-section, the secondary cortex is characterized by large subspherical groups of yellow sclereids and radially oriented medullary rays which are 1–2 cells broad. The parenchyma cells of the secondary cortex contain calcium oxalate prisms (up to 20 µm in length) and oil droplets. When viewed in longitudinal section, the sclereid aggregates are unusually long. The calcium oxalate crystals are arranged in longitudinal rows. Starch granules are infrequent, but when present, they are subspherical (2–6 µm in diameter). Scattered through the cortex are groups of stone cells and occasional isolated stone cells. Rifts or spaces of natural origin also occur in this region. Pericycle consists of parenchyma similar to the cortex, embedded with a few pericyclic fibres. The latter, when observed in longitudinal sections, possess an irregular lumen and obtuse or rounded ends. Phloem is of variable width, consisting of sieve tissue, phloem parenchyma and phloem rays; the phloem parenchyma contains starch grains, rosette aggregates and monoclinic prisms of calcium oxalate, tannin, amorphous orange, yellow or olive brown masses and oil. The phloem rays vary from straight to slightly curved or wavy and, while mostly 1 to 2 cells in width, may be up to 3 cells in width, as best determined from tangential-longitudinal sections through this region of the bark. Scattered through the phloem are numerous groups of stone cells. The margins of these groups are irregularly rounded, crenate, toothed and indented. Some of the phloem rays are intercepted in their outward course by groups of stone cells. As the stems grow older, secondary cork cambia originate successively in the cortex, pericycle and outer phloem forming wavy borke areas in these zones which contain groups of stone cells and, in the case of the pericyclic region, pericyclic fibres. In old stem bark, the medullary rays (phloem rays) reach outward to the periderm (*3*, *6*).

Root bark: cork of variable thickness, composed of somewhat lignified cork cells which are tangentially elongated in cross-sections and polygonal in surface sections, many of which possess orange or yellowish-orange or brownish contents. Phellogen of tangentially elongated meristematic cells. A secondary cortex of a narrow zone of tangentially elongated parenchyma cells, some of which contain orange or brownish amorphous masses, tannin, oil globules, starch, monoclinic prisms and rosette aggregates of calcium oxalate. Groups of stone cells with irregularly indented margins and isolated stone cells scattered through the region. Phloem consists of a relatively broad zone comprising a matrix of parenchyma and sieve tubes separated into a number of oblong or curved phloem patches by medullary rays which, in cross-sections, run nearly straight or curve and converge in groups. The stone cell groups are numerous and frequently deeply notched, the individual stone cells are strongly lignified with prominent

pore canals and rounded to irregular lumen, which frequently contain reddish to brownish content. Bast fibres absent. The medullary rays are mostly 1–2 cells wide, and contain either starch, tannin, orange-brownish amorphous masses or calcium oxalate crystals. Rosette aggregates of calcium oxalate found in the cells of the cortex, medullary rays and phloem are up to 54 µm in diameter. Monoclinic crystals also occur in these regions, but are fewer in number and only up to 25 µm in length. Starch grains present in these regions are simple and spheroidal to 2–3-compound, the individual grains being up to 23 µm in diameter (*6*).

Powdered plant material

Stem bark: light brown to moderate yellowish-brown. Cork layer in surface view. Stone cells numerous, rounded or elongated, in groups or isolated, with thick, porous, lignified walls and with reddish to brownish lumen and up to 260 µm in length; numerous fragments of lignified cork with brownish walls and polygonal in shape; calcium oxalate in rosette aggregates and monoclinic prisms up to 57 µm in diameter or length; fragments composed of parenchyma cells containing starch grains or calcium oxalate in rosette aggregates or monoclinic prisms, oil globules or orange to olive brown-coloured amorphous masses; starch grains simple or 2- to 3-compound, the individual grains spheroidal, ovate, elliptical, pyriform or plano-convex, up to 23 µm in diameter or length; a few fragments having wood fibres with lignified walls, some with lumen of irregular width and with bordered pores (*3*, *6*).

Root bark: to be established in accordance with national requirements.

General identity tests

Macroscopic and microscopic examinations (*3*, *6*) and thin-layer chromatography (*1*, *3*).

Purity tests

Microbiological

Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*7*).

Foreign organic matter

Not more than 3% (*1*).

Total ash

Not more than 12% (*1*).

Acid-insoluble ash
Not more than 4% (*1*).

Water-soluble extractive
Not less than 10% (*1*).

Alcohol-soluble extractive
To be established in accordance with national requirements.

Loss on drying
To be established in accordance with national requirements.

Pesticide residues
The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*8*). For other pesticides, see the *European pharmacopoeia* (*8*) and the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*7*) and pesticide residues (*9*).

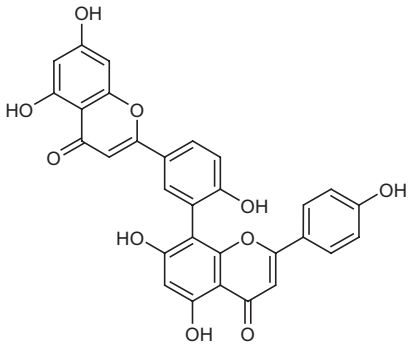
Heavy metals
For maximum limits and analysis of heavy metals, consult the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*7*).

Radioactive residues
Where applicable, consult the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*7*).

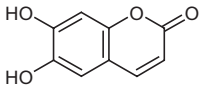
Chemical assays
To be established in accordance with national requirements.

Major chemical constituents
The chemical profile is somewhat diverse with iridoid glycosides (e.g. patrinoside, 2′-*O*-acetylpatrinoside and 2′-*O*-acetyl-dihydropenstemide), triterpenes (e.g. ursolic acid and ursolic acid acetate), coumarins (e.g. esculetin, scopoletin and scopolin) and the rarely found biflavonoid, amentoflavone, having been reported to be present in the crude drug (*3*, *5*). The structures of patrinoside, esculetin and amentoflavone are presented below.

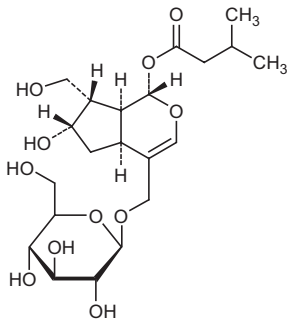
Medicinal uses
Uses supported by clinical data
No information was found.



Amentoflavone



Esculetin



Patrinoside

Uses described in pharmacopoeias and well established documents

Orally for the treatment of dysmenorrhoea and asthma (*1*, *10*, *11*).

Uses described in traditional medicine

Used to treat menstrual irregularities and nervous tension. Also used as an antispasmodic, diuretic and tonic (*5*).

Pharmacology

Experimental pharmacology

Antispasmodic activity

The data describing the antispasmodic effects of the crude drug are conflicting. Up until 1940, the plant material used was not properly identified and was thought to be commonly adulterated with other *Viburnum* species or even the inert mountain maple bark (*Acer spicatum*) (*12*). Thus, the lack of activity observed prior to 1940 may have been due to the use of adulterated plant materials.

Oral administration of a hot aqueous extract of the bark to pregnant dogs (50.0 ml/animal) or pregnant rabbits (25 ml/animal) had no inhibitory or stimulatory effects on uterine contractions (*13*). Intravenous administration of a 70% ethanol extract to cats at a dose of 8 ml/animal did not relax spontaneous uterine spasms (*13*). In addition, a fluidextract and hot aqueous extract were not active in vitro in tests in rat uteri, when assayed at a concentration of 1:1000 or 2:1000 (*14*).

Scientific research performed after 1940 with properly identified plant materials has yielded more positive results. In 1941, a modified version of the oxytocic assay from the 11th edition of the *United States Pharmacopeia*, in which the crude drug was tested for its ability to neutralize the oxytocic activity of the solution of posterior pituitary (*United States*

Pharmacopeia) was investigated. Addition of 0.1 ml of an ethanol extract of the bark to a 100-ml bath chamber containing isolated guinea-pig uterine strips, reduced normal and resting tone, and decreased the rate and amplitude of spontaneous contractions (*15*). The antispasmodic effects were observed 5–15 minutes after addition and when the extract was removed the contractions resumed. A synergistic effect was observed in rat uteri treated with the fluidextract of the bark and 1 mg papaverine hydrochloride (*16*). In a similar test, the essential oil of the bark reduced uterine contractions in isolated rat uteri (concentration not stated) (*17*).

An aqueous, 95% ethanol, methanol or fluidextract of the bark reduced spontaneous uterine contractions in the isolated uterus from female rats, at a concentration of 1:200, or 1mM (*11*, *18*, *19*). Methanol extracts of the bark had antispasmodic effects in estrone-primed barium chloride-stimulated rat uterine horns in vitro (*11*). Scopoletin, isolated from the bark had antispasmodic properties in vitro in estrone-primed and barium chloride-induced rat uteri. The concentration needed to produce a 50% decrease in the contraction amplitude of a single uterine horn was 0.09 mg/ml. Similar effects were observed in rodents in a study of oxytocin- and ergonovine-induced uterine contractions (*20*). A fluidextract of the bark (concentration not stated) reduced spontaneous contractions in rat uteri in vitro. Addition of a 95% ethanol extract (concentration not stated) of the bark to the bath medium prevented barium chloride and acetylcholine-induced spasms in isolated guinea-pig ileum (*21*).

A methanol extract (100 g dried bark), as well as the ethyl acetate and butanol fractions of the bark, inhibited spontaneous contractions of isolated rat uteri by 50% at concentrations of 2.0–200.0 µg/ml. A decrease in the tone and amplitude of acetylcholine-induced contractions was also observed. The median inhibitory concentration was 60 µg/ml for the crude extract, 36.0 µg/ml for the ethyl acetate fraction and 50 µg/ml for the butanol fraction. *2´-p*-Coumaroyldihydropenstemide was the active constituent (*22*).

Clinical pharmacology

No controlled clinical trials were found.

Adverse reactions

No information was found.

Contraindications

Cortex Viburni Prunifolii is contraindicated in cases of hypersensitivity or allergy to the crude drug.

Warnings

Use of the crude drug during pregnancy should only take place under the supervision of a health care professional.

Precautions

Carcinogenesis, mutagenesis, impairment of fertility

A 30% ethanol extract of the bark was not mutagenic in the Ames test at concentrations of 200 µl/disc in *Salmonella typhimurium* strains TA98 and TA100. Metabolic activation had no effect on the results (*23*).

Nursing mothers

No safety data are available, thus the use of the crude drug during breast-feeding is not recommended.

Paediatric use

No safety data are available, thus the use of the crude drug for children under the age of 12 years is not recommended.

Other precautions

No information was found.

Dosage forms

Crude drug, aqueous and ethanol extracts, fluidextracts and tinctures. Storage: protect from air, light, moisture, excessive heat and insect infestation (*3*).

Posology

(Unless otherwise indicated)

Powdered bark: 2.5–5 g as an infusion or decoction three times daily. Liquid extract (1:1) in 70% ethanol: dose 4–8 ml three times daily. Tincture (1:5 in 70% ethanol): 5–10 ml three times daily (*1*, *3*).

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Radix Withaniae

Definition
Radix Withaniae consists of the dried roots of *Withania somnifera* (L.) Dunal. (Solanaceae) (*1*, *2*).

Synonyms
Physalis somnifera L. (*3*).

Selected vernacular names
Achuvagandi, agol, ahan, aksin, amukkuram, amukkaramkizangu, amukkira, angarberu, a sh a ga n dha, asagand, asagandh, asagandh nagori, asagandha, asan, asana, askagandha as'vagandha, ashvagandha, ashvakandika, ashwaganda, ashwagandha, ashwaganha, asgand, asgandh, asgandha, asganhisrol, asoda, asun, asundha, asunyho, asuvagandi, asvagandha, asvagandhi, aswagandha, aswal, aswgandh, babu, bâibru, bouzidân, dambarico, ghoda, ghodakun, ghodasan, gisawa, gizawa, hayagandhã, hidi-budawa, hirchil, e-gaddy, hiremaddina-gaddy, hiremaddina-gida, Indian ginseng, juustumari, kakani hindi, kaknaj-e-hindi, kilangee, kuvia, lakri, ol asajet, oroval, penneru, pennerugadda, punir, samoah, sebbere-gola, sim-alfirakh, sum-ul-far, sum-ul-firakh, techil, ubab, u'beb, ubuvimba, vajigandha, winter cherry, withania (*1–7*).

Geographical distribution
Widespread from the Mediterranean coast to India in semi-arid habitats (*4*, *8*).

Description
A woody herb or shrub, up to 2 m in height; growing from a long, tuberous taproot; stellatetomentose. Leaf: simple, 2–11 cm in length by 1.5–9.0 cm in width, exstipulate, petiole 6–20 mm long; blade elliptic to ovate-lanceolate, apex acute or rounded, base acute to long-decurrent, on vegetative shoots 8–10 cm long and alternate, on reproductive shoots 3–8 cm long and opposite, arranged in pairs of one large and one smaller

leaf; margin entire or wavy. Inflorescence: axillary, umbellate cyme of 2–25 yellow-green, short-pedicellate flowers. Flower: perfect, radially symmetrical, campanulate; calyx with 5 acute triangular lobes; corolla twice the length of the calyx, 7–8 mm long, with 5 lanceolate lobes, spreading or reflexed; stamens 5, slightly exserted, filaments alternate to petal lobes, partially fused to corolla; ovary superior, glabrous, stigma shallowly bifid. Fruit: berry; globose, 5–6 mm in diameter, orange-red, enclosed in green, membranous, inflated calyx approximately 2.5 cm in diameter and slightly 5-angled. Seeds: many, discoid, 2.5 mm in diameter, pale yellow (*4*, *9*).

Plant material of interest: dried root

General appearance

Straight and unbranched, the thickness varying with age. The main roots bear fibre-like secondary roots. The outer surface of the root is buff to grey-yellow with longitudinal wrinkles. The crown consists of 2–6 remains of the stem base. The base of the stem is green, variously thickened, cylindrical and longitudinally wrinkled. The roots break with a short uneven fracture (*1*, *2*).

Organoleptic properties

Odour: characteristic, horse-like; taste: sweetish, yet bitter and astringent and slightly mucilaginous (*1*, *2*).

Microscopic characteristics

The transverse section shows a narrow band of yellowish cork, exfoliated or crushed, a narrow cortex packed with starch grains; cork cambium of 2–4 diffused rows of cells; secondary cortex about 24 layers of compact parenchymatous cells; phloem consists of sieve tube, companion cells, phloem parenchyma; cambium 4–5 rows of tangentially elongated cells; secondary xylem hard, forming a closed vascular ring separated by multiseriate medullary rays; a few xylem parenchyma (*1*, *2*).

Powdered plant material

Dusty white or grey to yellow-brown. Cork thin-walled; lignified, cubical or elongated cells, often indistinct and collapsed, with yellowish-brown contents; 2–3 cells deep in smaller roots, up to 16 in larger primary roots. Parenchyma of the cortex composed of large thin-walled cells, packed with starch granules, and occasionally containing microsphenoidal crystals of calcium oxalate. Xylem elements are either tracheidal with bordered pits or, more rarely, reticulately thickened vessels. Fibres from xy-

lem have thickened lignified walls and simple pits. Starch abundant, simple or 2–4-compound, with a pronounced irregularly shaped hilum (*4*, *9*).

General identity tests

Macroscopic and microscopic examinations (*1*), thin-layer chromatography (*4*) and high-performance liquid chromatography (*10*).

Purity tests

Microbiological

Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*11*).

Foreign organic matter

Not more than 2.0% (*1*, *2*).

Total ash

Not more than 7% (*1*, *2*).

Acid-insoluble ash

Not more than 1% (*1*, *2*).

Water-soluble extractive

To be established in accordance with national requirements.

Alcohol-soluble extractive

Not less than 15% (*1*, *2*).

Loss on drying

To be established in accordance with national requirements.

Pesticide residues

The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (*12*). For other pesticides, see the *European pharmacopoeia* (*12*) and the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*11*) and pesticide residues (*13*).

Heavy metals

For maximum limits and analysis of heavy metals, consult the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*11*).

Radioactive residues

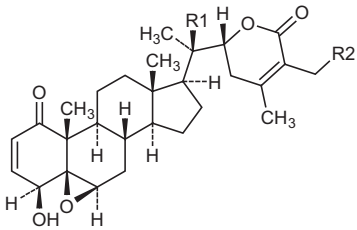
Where applicable, consult the WHO guidelines for assessing quality of herbal medicines with reference to contaminants and residues (*11*).

Chemical assays

Contains not less than 0.2% of total alkaloids determined by gravimetric method (*1*).

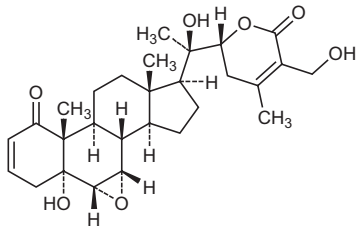
Major chemical constituents

The major characteristic constituents are steroidal lactones collectively known as "withanolides" including withaferin A, 27-deoxywithaferin A, withanolide D, withanosides I–XI, and withasomniferols A–C. Alkaloids constitute the other major group of compounds found in this plant material. Among the alkaloids found in the root are anaferine, anahygrine, cuscohygrine, dl-isopelletierine, 3-tropyltigloate, tropane-3-β-ol, 3-α-tigloyl-oxy-tropane and tropine. Also present are saponins including sitoindosides VII–X (*5*, *8*, *14*). The structures of withaferin A, withanolide D, withasomniferol A, cuscohygrine and dl-isopelletierine are presented below.

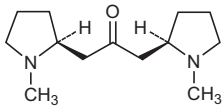


Withaferin A R1 = H R2 = OH

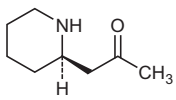
Withanolide D R1 = OH R2 = H



Withasomniferol A



Cuscohygrine



and enantiomer

Isopelletierine

Medicinal uses

Uses supported by clinical data

As an antistress agent to improve reaction time (*15*).

Uses described in pharmacopoeias and well established documents

As a general tonic to increase energy, improve overall health and prevent disease in athletes and the elderly (*1*, *16*).

Uses described in traditional medicine

Treatment of bronchitis, dyspepsia, impotency, scabies and ulcers (*5*, *16*).

Pharmacology

Experimental pharmacology

Antistress activity

The activity of a standardized extract of the root (1:1 aqueous ethanol fraction containing the withanolide glycosides and withaferin A at a concentration of 28–30%) was investigated in a rat model of chronic stress. A mild, unpredictable foot-shock was administered once daily for 21 days to rats. These chronic stress-induced perturbations were attenuated by the intragastric administration of the crude drug, at a dose of 25.0 and 50.0 mg/kg body weight (bw) given 1 h before foot-shock for 21 days (*17*).

In a mouse model of chronic stress, the antioxidant effects of the root were assessed in the forced swimming test. Biochemical analysis revealed that chronic swimming significantly increased lipid peroxidation and decreased glutathione levels in the brains of mice. The animal models also showed decreased levels of antioxidant defence enzymes, superoxide dismutase and catalase. Intragastric treatment with an extract of the crude drug, at a dose of 100.0 mg/kg bw significantly reduced lipid peroxidation and restored the glutathione levels decreased by chronic swimming in mice. Further, the treatment increased levels of superoxide dismutase in the forebrain and increased levels of catalase (*18*).

A withanolide-free aqueous fraction from the root (13 kg plant material in 70% ethanol, aqueous fraction) was evaluated for putative antistress activity. Intragastric administration of the preparation to immunocompromised mice for 7 days increased antibody production with a median effective dose of 40 mg/kg bw. Thus, the fraction exhibited significant antistress activity in a dose-related manner. The same fraction was protective against chemically and physically induced stress in rats and mice (*19*).

Intragastric administration of sitoindosides IX and X, at a dose range of 50–200.0 mg/kg bw also produced antistress activity in albino mice and rats, and augmented learning acquisition and memory retention in both young and old rats (*16*).

Anti-inflammatory activity

Numerous investigations have assessed the anti-inflammatory effects of the crude drug in vitro and in vivo (*20–22*). In one study, a suspension of powdered root (1 g/kg suspended in 2% acacia gum, 50 mg/ml) administered intragastrically to rats for 3 days, 1 hour before the injection of Freund's complete adjuvant, reduced inflammation. Many serum proteins such as α 2-glycoprotein, a major acute inflammatory phase protein and pre-albumin were decreased, indicating a reduction in acute inflammation (*20*). In another study by the same research group, 1 g/kg of the root suspension reduced the α 2-macroglobulin in the serum of rats given subplantar injection of carrageenan suspension (*21*).

In one study, air pouch granuloma was induced by subcutaneous injections of 4 ml of carrageenan on the dorsum of rats which had been subcutaneously injected 1 day previously with 6 ml of air on the dorsum (*22*). The powdered crude drug was administered by gastric lavage at a dose of 1 g/kg bw for 3 days. Radioactive sodium sulfate was injected intraperitoneally on day 9 and the incorporation of radio-labelled sulfur in glycosaminoglycan, oxidative phosphorylation (ADP/O ratio), mg^{2+}-dependent-ATPase enzyme activity and succinate dehydrogenase activity were determined in the mitochondria of the granuloma tissue. Administration of the root decreased the glycosaminoglycan content of the granuloma tissue by 92%, compared with 43.6% following treatment with hydrocortisone (15.0 mg/kg bw) and had no effect following treatment with phenylbutazone (100.0 mg/kg bw) (*22*).

In a further study, the effect of oral administration of the crude drug (root powder, 1 g/kg bw, daily for 15 days) on paw swelling and bony degenerative changes in Freund's complete adjuvant-induced arthritis in rats was assessed. Intragastric administration of the powdered root to the rats caused a significant reduction in both paw swelling and degenerative changes as observed by radiological examination. The reductions were better than those produced by the reference drug, hydrocortisone (15.0 mg/kg bw) (*16*, *23*).

Daily intragastric administration of 1.0 g/kg bw of the powdered root suspended in 2% gum acacia, was given 1 hour before the induction of inflammation by injection of Freund's complete adjuvant in rats for 3 days. Phenylbutazone (100.0 mg/kg bw) was administered to another group of animals as a positive control. Assessment of acute phase reactants of the blood showed changes in the concentration of α 2-glycoprotein, major acute phase α 1-protein, and pre-albumin in rats with inflammation, while in animals treated with the powdered root, the α 2-

glycoprotein and acute phase protein were decreased to undetectable levels (*16*).

Intragastric administration of the root, at doses of 500, 1000, 1500 or 1200 mg/kg bw, given orally as a suspension 3–4 hours prior to induction of inflammation, also caused a dose-dependent suppression of α 2-macroglobulin (an indicator for anti-inflammatory drugs) in the serum of rats inflamed by subplantar injection of carrageenan. The maximum effect (about 75%) was seen at 1.0 g/kg bw. Actual measurements of inflammation were not made (*16*). Glycowithanolides and a mixture of sitoindosides IX and X isolated from the crude drug were evaluated for their immunomodulatory and central nervous system effects (antistress and improvements to memory and learning) in mice and rats. Both mixtures of compounds led to mobilization and activation of peritoneal macrophages, phagocytosis and increased activity of the lysosomal enzymes (*16*).

Anti-ischaemic activity

An in vivo study assessed the effect of the crude drug as a prophylactic treatment against stroke using the middle cerebral artery occlusion model in rats. Two groups of rats were pretreated with a hydroalcoholic extract of the crude drug (1.0 g/kg, administered by gastric lavage) for 15 or 30 days. The rats were then subjected to focal ischaemia by occlusion of the middle cerebral artery using an intraluminal thread. After 2 hours of middle cerebral artery occlusion, reperfusion was allowed by retracting the thread and the animals were assessed for ischaemic changes 30 min after reperfusion. Twenty-four hours later, rats were subjected to motor performance tests and were subsequently killed to enable estimation of the marker of oxidative stress, malondialdehyde. Significant motor impairment, with elevated levels of malondialdehyde, was observed in middle cerebral artery-occluded control rats. Treatment with the crude drug for 30 days prevented motor impairment and significantly decreased the raised levels of malondialdehyde compared with rats treated with the vehicle. Treatment also attenuated the percentage hemispheric lesion area in diffusion-weighted imaging (17 ± 2%) compared with the vehicle-treated middle cerebral artery-occluded group (30 ± 4%) (*24*).

A study was conducted to evaluate the cardioprotective potential of a hydro-alcoholic extract of the roots, by assessing the effects of treatment on haemodynamic, histopathological and biochemical parameters in isoprenaline- (isoproterenol-) induced myocardial necrosis in rats and to compare the effects with those of vitamin E (*25*). Rats were divided into six main groups: sham, isoprenaline control, *Withania somnifera* and vitamin E controls and *Withania somnifera* and vitamin E treatment groups. The root extract was administered at doses of 25, 50 and 100 mg/kg, and

vitamin E at a dose of 100 mg/kg, orally for 4 weeks. On days 29 and 30, the rats in the isoprenaline control group and the *Withania somnifera* and vitamin E treatment groups were given isoprenaline (85 mg/kg), subcutaneously at an interval of 24 hours. On day 31, haemodynamic parameters were recorded before the animals were killed, and the hearts were subsequently removed and subjected to histopathological and biochemical studies. A significant decrease in glutathione ($p < 0.05$), activities of superoxide dismutase, catalase, creatinine phosphokinase and lactate dehydrogenase ($p < 0.01$) as well as an increase in the level of the lipid peroxidation marker malonyldialdehyde ($p < 0.01$) was observed in the hearts of rats in the isoproterenol control group as compared to rats in the sham control group. Treatment with the root extract exerted a strong protective effect against isoprenaline-induced myonecrosis in rats. The dose of 50 mg/kg bw of the root extract had the greatest cardioprotective effect of the treatments studied (*25*).

Antioxidant activity

Dried ethanol extracts of the roots were tested for their total antioxidant activity in the iron (Fe^{3+}) reducing assay, and found to be potent reductants of Fe^{3+} at pH 5.5 (*26*). In an in vivo study, the powdered roots were assessed for their ability to protect neurons against excitotoxic lesions induced by kainic acid in mice. Mice were anaesthetized with ketamine and xylazine and kainic acid was administered by intra-hippocampal injections. The results showed an impairment of the function of the hippocampus region of brain after injection of kainic acid, and lipid peroxidation and protein carbonyl content were significantly increased in comparison with control animals ($p < 0.05$). The extract given 3 weeks prior to injections of kainic acid resulted in a decrease in neurotoxicity and measures of lipid peroxidation and protein carbonyl declined. The results of this study suggest that the crude drug mitigates the effects of excitotoxicity and oxidative damage in hippocampus by its antioxidative properties (*26*).

The glycowithanolides isolated from the crude drug were investigated for their preventive effect on the animal model of tardive dyskinesia, induced by once daily administration of the neuroleptic, haloperidol, for 28 days. Involuntary orofacial movements chewing movements, tongue protrusion and buccal tremors were assessed as parameters of tardive dyskinesia. Intragastric administration of 100.0 and 200.0 mg of the root, concomitantly with haloperidol, for 28 days inhibited the induction of the neuroleptic tardive dyskinesia (*27*).

The effects of the roots against oxidative stress in haloperidol-induced orofacial dyskinesia (haloperidol-induced vacuous chewing movements and tongue protrusion) were assessed in rats. Animals were treated for 21 days with intraperitoneal haloperidol (1 mg/kg); on day 22, vacuous chewing movements and tongue protrusions were counted during a 5-minute observation period. Coadministration of the extract (100–300 mg/kg bw) dose-dependently reduced haloperidol-orofacial dyskinesia. Biochemical analysis revealed that chronic treatment with haloperidol significantly increased lipid peroxidation and decreased forebrain levels of glutathione and the antioxidant defence enzymes, superoxide dismutase and catalase. Coadministration of the crude drug extract significantly reduced the lipid peroxidation and significantly reversed the decrease in forebrain superoxide dismutase and catalase levels, but had no significant effect on the haloperidol-induced decrease in forebrain glutathione levels (*28*).

The sitoindosides VII–X and withaferin A (glycowithanolide), were tested for antioxidant activity using the major free-radical scavenging enzymes, superoxide dismutase, catalase and glutathione peroxidase levels in the rat brain frontal cortex and striatum. Active glycowithanolides of *Withania somnifera* (10.0 or 20.0 mg/kg bw) were administered by intraperitoneal injection once daily for 21 days to groups of six rats. Dose-related increases in all enzymes were observed; the increases were comparable to those seen following administration of deprenyl (2 g/kg bw intraperitoneally), indicating that the crude drug has an antioxidant effect in the brain which may be responsible for its diverse pharmacological properties (*29*).

In another study, an aqueous suspension of the crude drug was evaluated for its effect on stress-induced lipid peroxidation in mice and rabbits (*30*). Levels of lipid peroxidation in the blood were increased by intravenous administration of lipopolysaccharides from *Klebsiella pneumoniae* and peptidoglycans from *Staphylococcus aureus*. Simultaneous intragastric administration of the extract (100.0 mg/kg bw) prevented an increase in lipid peroxidation.

In another study, the powdered root was administered to mice at a dose of 0.7 and 1.4 g/kg bw, for 15 and 30 days, to determine its effects on lipid peroxidation, superoxide dismutase and catalase activities. Thirty days of treatment produced a significant decrease in lipid peroxidation, and an increase in both superoxide dismutase and catalase activities (*31*). The effect of an extract of the crude drug on the regulation of lead toxicity and lipid peroxidative activity was investigated in liver and kidney tissues of rodents. Lead treatment of the animals for 20 days enhanced

hepatic and renal lipid peroxidation, whereas administration of the extract at doses of 0.7 g/kg bw and 1.4 g/kg bw together with equivalent doses of lead acetate for 20 days significantly decreased lipid peroxidation and increased the activities of antioxidant enzymes, such as superoxide dismutase and catalase (*32*).

An in vivo study examined the attenuating effect of extracts of the root and of aloe vera on prevention of hippocampal and cortical cell degeneration due to oxidative damage in mice with streptozotocin-induced diabetes. Doses of both plant extracts given to experimental animals were based on the evaluation of their total antioxidant activity and also their potency to reduce Fe^{3+}. Lipid peroxidation and protein carbonyl were assayed in both regions of the brain and the changes in memory and motor behavioural functions in diabetic and control mice were observed. The results showed a significant increase in lipid peroxidation and protein carbonyl in the hippocampus and cortical regions of mice with streptozotocin-induced diabetes, as well as a significant impairment in both motor and memory behavioural functions in diabetic mice. However, when diabetic mice were supplemented with the extracts of the root and with aloe vera, the oxidative damage in both brain regions was reduced as marked by a significant decline in both lipid peroxidation and protein carbonyl. The combination of extracts of root and aloe vera was more effective in reducing oxidative damage in brain regions than either of the plant extracts given alone. The combination of the extract and the aloe vera lowered the blood glucose level in mice with streptozotocin-induced diabetes. Memory impairment and motor dysfunction were also lessened by supplementation with the plant extracts (*33*).

Chemopreventive activity

The chemopreventive effect of an alcohol extract of the crude drug on 7,12-dimethylbenz[a]anthracene (DMBA)-induced skin cancer was investigated in mice. The skin lesions were induced by the twice-weekly topical application of DMBA for 8 weeks to the shaved backs of mice. The alcohol extract was administered at the maximum oral tolerated dose of 400.0 mg/kg bw three times per week on alternate days 1 week before DMBA application and treatment was continued for 24 weeks. The results showed a significant decrease in incidence and average number of skin lesions in mice that received the extract compared to those treated with DMBA alone at the end of week 24. A significant impairment was noticed in the levels of reduced glutathione, malondialdehyde, superoxide dismutase, catalase, glutathione peroxidase and glutathione S-transferase in skin lesions of DMBA-treated control mice compared with vehicle-

treated mice. These parameters returned to near normal following administration of the crude drug to DMBA-treated mice (*34*).

Another chemopreventive study of an alcohol extract of the crude drug against 20-methylcholanthrene-induced fibrosarcoma tumours in mice was performed. A single subcutaneous injection of 20-methylcholanthrene into the thigh region of the mice produced a high incidence (96%) of tumours. Intragastric administration of the extract at a dose of 400.0 mg/kg bw (1 week before injecting 20-methylcholanthrene and continued until 15 weeks thereafter) significantly reduced the incidence and volume of tumours and enhanced the survival of the mice, compared with mice injected with 20-methylcholanthrene. The tumour incidence was also delayed in the group treated with the extract when compared with mice injected with 20-methylcholanthrene without pretreatment with the extract. Liver biochemical parameters revealed a significant modulation of reduced glutathione, lipid peroxides, glutathione S-transferase, catalase and superoxide dismutase in mice treated with the extract compared with mice injected with 20-methylcholanthrene (*35*).

Effects on memory and cognition

An aqueous suspension (100 mg/ml of powdered root suspended in 2% acacia gum and water) was assessed for its effects on improving short-term memory by reversing the effects of memory deficits induced by scopolamine and amnesia induced by electroconvulsive shock (*36*). Daily administration of 200 mg/kg bw of the suspension significantly reversed the scopolamine-induced delay in latency for the animals to reach the shock free zone and the number of errors in the passive avoidance step-down test ($p < 0.001$). Treatment of the animals with 100 mg/kg bw of the suspension produced a significant reduction in latency to reach the shock free zone ($p < 0.05$) (*36*).

Sitoindosides VII–X and withaferin A were isolated from an aqueous methanol extract of the roots. Rats were treated with 40 mg/kg bw (intraperitoneally) of an equimolar mixture of these compounds for 7 days and the anticholinesterase activity was determined in brain slices. Acetylcholinesterase activity was increased in the lateral septum and globus pallidus indicating a possible enhancement of cognition (*37*).

Withanolides isolated from the crude drug inhibited acetylcholinesterase and butyrylcholinesterase enzymes in a concentration-dependent fashion with IC_{50} values ranging between 20.5 and 85.2 μM for acetylcholinesterase and butyrylcholinesterase. Lineweaver-Burk as well as Dixon plots and their secondary replots indicated that the compounds were linear mixed-type inhibitors of acetylcholinesterase and non-competitive inhibitors of acetylcholinesterase with K_i (dissociation constant) values

ranging between 20.0 and 45.0 μM. All compounds were found to be non-competitive inhibitors of butyrylcholinesterase with K_i values ranging between 27.7 and 90.6 μM (*38*).

Immune stimulant activity

The effects of the root were investigated in mice with myelosuppression induced by cyclophosphamide, azathioprin or prednisolone (*39*). Administration of the root (100 mg/kg bw) prevented myelosuppression in mice treated with all three immunosuppressive drugs tested. A significant increase in haemoglobin concentration ($p < 0.01$), red blood cell count ($p < 0.01$), white blood cell count ($p < 0.05$), platelet count ($p < 0.01$), and body weight ($p < 0.05$) was observed in treated mice as compared with untreated (control) mice (*39*).

The effect of the crude drug on the cellular immune responses was studied in normal and in tumour-bearing animals. Intraperitoneal injection of five doses of an extract of the crude drug (20 mg/dose/animal) enhanced the proliferation of lymphocytes, bone marrow cells and thymocytes in response to mitogens. Both phytohaemagglutinin and concanavalin A mitogens administered concomitantly with crude drug treatment doubled the proliferation of lymphocytes, bone marrow cells and thymocytes. Splenocytes treated with the crude drug together with the mitogen led to a six-fold increase in lymphocyte proliferation. Natural killer cell activity was stimulated by the crude drug extract in both normal and tumour-bearing animals and it was found to be earlier than in the controls (48.92% cell lysis). Antibody-dependent cellular cytotoxicity was found to be enhanced in the group treated with the crude drug on the ninth day after treatment (65% cell lysis) (*40*). These authors have also reported that intraperitoneal administration of a 70% methanol extract of the roots to mice at a dose of 20 mg/animal enhanced total white blood cell count on day 10 after the administration of a single dose; bone marrow cellularity also increased, and the delayed-type hypersensitivity reaction (Mantoux test) was reduced (*41*).

The immunomodulatory effects of extracts of the crude drug (suspensions in carboxymethyl cellulose) were investigated in mice to measure their effects on immune hyper-reactivity. The animal models used included antibody-mediated immune hyper-reactivity, as seen in active paw anaphylaxis with disodium chromoglycate and cell-mediated immune hyper-reactivity using the delayed-type hypersensitivity model with cyclophosphamide. The immunomodulatory effect was assessed in IgE-mediated anaphylaxis as the reduction of ovalbumin-induced paw oedema in animals treated with the crude drug suspension at doses of 150 and

300.0 mg/kg bw. The positive control drug used was disodium chromoglycate. Potentiation of the delayed-type hypersensitivity reaction was observed in animals treated with cyclophosphamide at a dose of 20.0 mg/kg bw, and the crude drug suspensions at a dose of 300–1000.0 mg/kg bw. A significant increase in white blood cell counts and platelet counts was observed in animals treated with the crude drug. A protective effect against cyclophosphamide-induced myelosuppression was observed in animals treated with suspensions of the crude drug at doses of 300–1000.0 mg/kg bw, and a significant increase in white blood cell counts and platelet counts was observed. Cyclophosphamide-induced immunosuppression was counteracted by treatment with the same crude drug suspension. Treated animals showed an increase in haemagglutinating antibody responses and haemolytic antibody responses towards sheep red blood cells (*42*).

In a study in mice, intraperitoneal administration of a methanol extract of the root (20.0 mg/kg bw) was found to significantly reduce leukopaenia induced by treatment with cyclophosphamide. On the twelfth day the total white blood cell count in the group treated with cyclophosphamide was 3720 cells/mm^2 and that of the group that received cyclophosphamide together with the root was 6120 cells/mm^2. Treatment with the root and cyclophosphamide significantly increased the bone marrow cellularity (13.1×10^6 cells/femur) compared to the group treated with cyclophosphamide alone (8×10^6 cells/femur) ($p < 0.001$). Administration of the extract increased the number of alpha-esterase positive cells in the bone marrow of animals treated with cyclophosphamide, compared with the animals in the group treated with cyclophosphamide alone (687/4000 cells) (*43*).

The mechanism underlying the immunostimulant effect of a methanol extract of the root was investigated by assessing nitric oxide production in J774 macrophages (*44*). At concentrations of 1–256 µg/ml the extract produced a significant and concentration-dependent increase in nitric oxide production, an effect which was abolished by N(G)nitro-L-arginine methyl ester, a non-selective inhibitor of nitric oxide synthase; dexamethasone, an inhibitor of protein synthesis; and N(alpha-p)-tosyl-L-lysine chloromethyl ketone, an inhibitor of nuclear factor-kappa-β activation. In addition, Western blot analysis showed that the methanol extract increased, in a concentration-dependent fashion, expression of inducible nitric oxide synthase protein. The results suggest that the crude drug may induce the synthesis of inducible nitric oxide synthase expression, probably by acting at the transcriptional level, and the increased nitric oxide production by macrophages could account, at least in part, for the immunostimulant properties (*44*).

Neuroprotective activity

Eighteen compounds isolated from a methanol extract of the crude drug enhanced neurite outgrowth in human neuroblastoma cells. Double immunostaining was performed in rat cortical neurons using antibodies to phosphorylated nuclear factor-H as an axonal marker, and to mitogen activated protein kinase as a dendritic marker. In cells treated with withanolide A, the length of nuclear factor-H-positive processes was significantly increased compared with that of vehicle-treated cells, whereas the length of mitogen activated protein kinase-positive processes was increased by withanosides IV and VI. These results suggest that axons are predominantly extended by withanolide A, and dendrites by withanosides IV and VI (*45*).

The anti-parkinsonian effects of a root extract were investigated in rats pretreated with 100, 200 and 300 mg/kg bw of the root extract orally for 3 weeks. On day 21, 2 µl of 6-hydroxydopamine was infused into the right striatum while animals in the sham-operated group received 2 µl of the vehicle solvent. Three weeks after being injected with 6-hydroxydopamine, the rats were tested for neurobehavioural activity and 5 weeks after treatment they were killed for the estimation of lipid peroxidation, reduced glutathione content, activities of glutathione S-transferase, glutathione reductase, glutathione peroxidase, superoxide dismutase and catalase, catecholamine content, dopaminergic D2-receptor binding and tyrosine hydroxylase expression. The root extract reversed all the parameters in a dose-dependent manner indicating a potential for preventing neuronal injury in Parkinson disease (*46*).

Toxicity

In one study of its effects on the central nervous system, a 2% aqueous suspension of the powdered root, an alkaloid-containing extract of the root prepared in 10% propylene glycol using 2% gum acacia as the suspending agent, was used to determine acute toxicity (*47*). The acute median lethal dose (LD_{50}) was 465.0 mg/kg bw in rats and 432.0 mg/kg bw in mice. In an antistress-effect study, the acute toxicity of aqueous-methanol extracts of the crude drug and equimolar combinations of sitoindosides VII and VIII (SG-I) and withaferin-A (SG-2) were studied. The acute LD_{50} of SG-1 and SG-2 administered intraperitoneally to mice was 1076.0 mg/kg bw and 1564.0 mg/kg bw, respectively (*48*).

In one study of the effects of chronic administration, the root was boiled in water and administered to rats in their drinking-water for 8 months while monitoring body weight, general toxicity, well-being, number of pregnancies, litter size and weight of progeny (*16*). The estimated dose given was 100.0 mg/kg bw per day. In the second part of

the study, an estimated dose of 200 mg/kg bw per day was given for 4 weeks as above while monitoring body temperature, body weight, cortisol value in heparinized plasma and ascorbic acid content of the adrenals. The liver, spleen, lungs, kidneys, thymus, adrenals and stomach were examined histopathologically and were all found to be normal. The initial average body weights of the animals in the group treated with the crude drug (100.0 mg/kg bw per day) and in the control group on day 1 were 91 g and 106 g, which, after 4 weeks, had increased to 185 g (103%) and 178 g (67.9%), respectively. The rats treated with the crude drug gained more weight than those in the control group (no p value given). The percentage weight gain after 8 weeks of the same *Withania somnifera* treatment was 227% for the animals in the treated group and 145.3% for those in the control group. The relative body weight gain was significantly greater in the group treated with the crude drug than in the control group ($p < 0.001$). While it is not clear when the rats were mated, the average weights of the progeny at 1 month of age were 70 g and 45 g in the crude-drug-treated and control groups, respectively, indicating healthier progeny in the treated group. In the 4-week study, the weight gain in the animals in the treated group was comparable to that of those in the control group. The body temperature of animals in the group treated with the crude drug was 1.7 °C lower than in the control animals. The treatment caused an increase in lung and liver weights and a decrease in adrenocortical activity as was evident from the reduction in adrenal weight and a significant reduction in plasma cortisol ($p < 0.001$). Histopathologically, all organs were normal, and no toxicity was observed (*16*).

Clinical pharmacology

A double-blind, placebo-controlled clinical trial assessed the effects of the root (250 mg twice daily) on psychomotor performance in 30 healthy volunteers (*15*). The effects were compared with those of *Panax ginseng* (100 mg twice daily). Test parameters included tapping, cancellation test, mental mathematical calculations, logical deductions, choice reaction times and auditory reactions. The performance of both groups was superior to that of subjects who received a placebo and the performance of subjects given the crude drug was superior to that of those given *Panax ginseng* after 40 days of treatment.

Adverse reactions

May cause nausea, vomiting and diarrhoea (*4*).

Contraindications
Due to the lack of safety data and the fact that the crude drug has been used in traditional medicine to induce abortion, its use during pregnancy or breastfeeding is contraindicated (*4*).

Warnings
No information was found.

Precautions
Drug interactions
The crude drug may potentiate the effects of barbiturates and reduce the effects of diazepam and clonazepam (*4*).

Carcinogenesis, mutagenesis, impairment of fertility
No information was found.

Pregnancy: non-teratogenic effects
See Contraindications.

Other precautions
No information was found.

Dosage forms
Crude drug, extracts and tinctures.

Posology
(Unless otherwise indicated)
Powdered crude drug: 3–6 g of the dried powdered root (*1*). Orally as an antistress agent: 250 mg twice daily (*15*).

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Annex 1

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Annex 2

Cumulative index
(*in alphabetical order of plant name*)

For the convenience of users of Volume 4, the monographs described in Volumes 1, 2, and 3 are also listed in this index. The numbers printed in bold type, preceding the page numbers, indicate the volume in which the indexed item is to be found. Monographs are listed in alphabetical order of the plant name.

Annex 3

Cumulative index

(*in alphabetical order of plant material of interest*)

For the convenience of users of Volume 4, the monographs described in Volumes 1, 2 and 3 are also listed in this index. The numbers printed in bold type, preceding the page numbers, indicate the volume number in which the indexed item is to be found. Monographs are listed in alphabetical order of the plant material of interest.

Annex 4

Cumulative index of medicinal plants (*in alphabetical order of Latin binomial plant name*)

For the convenience of users of Volume 4, the medicinal plants described in Volumes 1, 2, and 3 are also listed in this index. The plants are listed in alphabetical order of their Latin binomial name along with the family they belong to in square brackets. The specific monograph(s) in which the plant is defined is listed next to the white bullet. The numbers printed in bold type, preceding the page numbers, indicate the volume number in which the monograph is found.

Annex 5

Cumulative index of major chemical constituents (*by compound name in alphabetical order*)

Cumulative index of major chemical compounds whose chemical structure drawings were presented in four volumes of WHO monographs on selected medicinal plants

Names are classified in alphabetical order. Some of the names are considered as synonyms, they are followed by the name of the "leading name".
CAS RN is the number of the compound in the *Chemical Abstracts Service* database.
Mol. form. is the molecular formula.
Volume number (in bold) of the *WHO monographs on selected medicinal plants* is given followed by __ and the **page** within the volume, where one can find the structure of the compound.
The corresponding **monograph name** is given in the last column.

Name	CAS RN	Mol. form.	Volume	Monograph name
acetoxyvalerenic acid	84638-55-1	C-17 H-24 O-4	**1**_271	Radix **V**alerianae
3-*O*-acetyl-11-oxo-β-boswellic acid	67416-61-9	C-32 H-48 O-5	**4**_51	Gummi **B**oswellii
3-*O*-acetyl-β-boswellic acid	5968-70-7	C-32 H-50 O-4	**4**_51	Gummi **B**oswellii
3-*O*-acetyl-α-boswellic acid	89913-60-0	C-32 H-50 O-4	**4**_51	Gummi **B**oswellii
4-acetylguaiacol (see apocynine)				
2′-*O*-acetylsalicortin		C-22 H-26 O-11	**4**_312	Cortex **S**alicis
acevaltrate	25161-41-5	C-24 H-32 O-10	**1**_271	Radix **V**alerianae
achillicin	71616-00-7	C-17 H-22 O-5	**4**_184	Herba **M**illefolii
acintene A (see α-pinene)				
actein	18642-44-9	C-37 H-56 O-11	**2**_58	Rhizoma **C**imicifugae Racemosae
acteol	27208-74-8	C-30 H-46 O-6	**2**_58	Rhizoma **C**imicifugae Racemosae
adhyperforin	143183-63-5	C-36 H-54 O-4	**2**_153	Herba **H**yperici

Name	CAS RN	Mol. form.	Volume	Monograph name
aesculetin (see esculetin)				
agnuside	11027-63-7	C-22 H-26 O-11	**4**_13	Fructus **A**gni Casti
ailanthinone	53683-73-1	C-25 H-34 O-9	**1**_62	Fructus **B**ruceae
(*E*)-ajoene	92284-99-6	C-9 H-14 O S-3	**1**_19	Bulbus **A**lli Sativi
(*Z*)-ajoene	92285-00-2	C-9 H-14 O S-3	**1**_19	Bulbus **A**lli Sativi
ajugol	52949-83-4	C-15 H-24 O-9	**3**_286	Radix **R**ehmanniae
alizarine yellow (see ellagic acid)				
allicin	539-86-6	C-6 H-10 O S-2	**1**_19	Bulbus **A**lli Sativi
alliin	556-27-4	C-6 H-11 N O-3 S	**1**_19	Bulbus **A**lli Sativi
aloe-emodin	481-72-1	C-15 H-10 O-5	**1**_234	Rhizoma **R**hei
aloin A	1415-73-2	C-21 H-22 O-9	**1**_37;	**A**loe
			2_262	Cortex **R**hamni Purshianae
aloin B	28371-16-6	C-21 H-22 O-9	**1**_37;	**A**loe
			2_262	Cortex **R**hamni Purshianae
aloinoside A	56645-88-6	C-27 H-32 O-13	**1**_37	**A**loe
aloinoside B	11006-91-0	C-27 H-32 O-13	**1**_37	**A**loe
alphanon (see camphor)				
alphitolic acid	19533-92-7	C-30 H-48 O-4	**3**_362	Fructus **Z**izyphi
amarine (see cucurbitacin B)				
amarogentin	21018-84-8	C-29 H-30 O-13	**3**_153;	Radix **G**entianae Luteae
			3_162	Radix **G**entianae Scabrae
amentoflavone	1617-53-4	C-30 H-18 O-10	**1**_157;	Folium **G**inkgo
			4_369	Cortex **V**iburni Prunifolii
ammidin (see imperatorin)				
ammiol	668-10-0	C-14 H-12 O-6	**3**_26	Fructus **A**mmi Visnagae
ammoidin (see xanthotoxin)				
amygdalin	29883-15-6	C-21 H-27 N O-11	**3**_66	Semen **A**rmenicae
α-amyrin	638-95-9	C-30 H-50 O	**2**_272	Flos **S**ambuci
andrographiside	82209-76-5	C-26 H-40 O-10	**2**_15	Herba **A**ndrographidis
andrographolide	5508-58-7	C-20 H-30 O-5	**2**_15	Herba **A**ndrographidis
andropanoside	82209-72-1	C-26 H-40 O-9	**2**_15	Herba **A**ndrographidis
trans-anethole	4180-23-8	C-10 H-12 O	**3**_44;	Aetheroleum **A**nisi
			3_56;	Fructus **A**nisi
			3_139	Fructus **F**oeniculi
angelicide	92935-94-9	C-24 H-28 O-4	**2**_28	Radix **A**ngelicae Sinensis

Name	CAS RN	Mol. form.	Volume	Monograph name
angelicone	37719-98-5	C-16 H-16 O-5	**2**_28	Radix **A**ngelicae Sinensis
angelol G	83199-38-6	C-20 H-24 O-7	**2**_28	Radix **A**ngelicae Sinensis
p-anisaldehyde	123-11-5	C-8 H-8 O-2	**3**_44;	Aetheroleum **A**nisi
			3_56;	Fructus **A**nisi
			3_139	Fructus **F**oeniculi
apigenin	520-36-5	C-15 H-10 O-5	**1**_89	Flos **C**hamomillae
apocynine	498-02-2	C-9 H-10 O-3	**4**_262	Rhizoma **P**icrorhizae
arasaponin E1 (see ginsenoside Rb1)				
arbutin	497-76-7	C-12 H-16 O-7	**2**_345	Folium **U**vae Ursi
artecanin	29431-84-3	C-15 H-18 O-5	**2**_320	Herba **T**anaceti Parthenii
asiatic acid	464-92-6	C-30 H-48 O-5	**1**_80	Herba **C**entellae
asiaticoside	16830-15-2	C-48 H-78 O-19	**1**_80	Herba **C**entellae
asterin (see cyanidin 3-monoglucoside)				
astragalin	480-10-4	C-21 H-20 O-11	**2**_39	Flos **C**alendulae
astragaloglucans			**1**_53	Radix **A**stragali
astragaloside I	84680-75-1	C-45 H-72 O-16	**1**_53	Radix **A**stragali
astragaloside II	84676-89-1	C-43 H-70 O-15	**1**_53	Radix **A**stragali
astragaloside III	84687-42-3	C-41 H-68 O-14	**1**_53	Radix **A**stragali
astragaloside IV	84687-43-4	C-41 H-68 O-14	**1**_53	Radix **A**stragali
astragaloside V	84687-44-5	C-47 H-78 O-19	**1**_53	Radix **A**stragali
astragaloside VI	84687-45-6	C-47 H-78 O-19	**1**_53	Radix **A**stragali
astragaloside VII	84687-46-7	C-47 H-78 O-19	**1**_53	Radix **A**stragali
aucubin	479-98-1	C-15 H-22 O-9	**3**_286;	Radix **R**ehmanniae
			4_13;	Fructus **A**gni Casti
			4_262	Rhizoma **P**icrorhizae
aucubin *p*-hydroxybenzoate (see agnuside)				
azadirachtin	11141-17-6	C-35 H-44 O-16	**3**_91;	Folium **A**zadirachti
			3_104	Oleum **A**zadirachti
baicalein	491-67-8	C-15 H-10 O-5	**3**_316	Radix **S**cutellariae
baicalin	21967-41-9	C-21 H-18 O-11	**3**_316	Radix **S**cutellariae
baichanin A (see genistein)				
baldrinal	18234-46-3	C-12 H-10 O-4	**1**_271	Radix **V**alerianae
barbaloin (see aloin)				
barringtogenol C	13844-01-4	C-30 H-50 O-5	**2**_140	Semen **H**ippocastani
bellaradine (see cuscohygrine)				
benzoaric acid (see ellagic acid)				
berbamine	478-61-5	C-37 H-40 N-2 O-6	**4**_33	Cortex **B**erberidis
berbericinine (see palmatine)				
berberine	2086-83-1	C-20 H-18 N O-4	**1**_109;	Rhizoma **C**optidis
			3_197;	Rhizoma **H**ydrastis

Name	CAS RN	Mol. form.	Volume	Monograph name
			4_33;	Cortex **B**erberidis
			4_247	Cortex **P**hellodendron
bergapten	484-20-8	C-12 H-8 O-4	**3**_12	Fructus **A**mmi Majoris
betulinic acid	472-15-1	C-30 H-48 O-3	**3**_362	Fructus **Z**izyphi
I3´,II8-biapigenin (see amentoflavone)				
2,2´-bichavicol (see magnolol)				
bilobalide	33570-04-6	C-14 H-16 O-8	**1**_157	Folium **G**inkgo
bilobetin	521-32-4	C-31 H-20 O-10	**1**_157	Folium **G**inkgo
biochanin A	491-80-5	C-16 H-12 O-5	**4**_338	Flos **T**rifolii
biochanin B (see formononetin)				
(-)-α-bisabolol	23089-26-1	C-15 H-26 O	**1**_89	Flos **C**hamomillae
bisdesmethoxycurcumin	33171-05-0	C-19 H-16 O-4	**1**_118	Rhizoma **C**urcumae Longae
2-bornanone (see camphor)				
borneol	507-70-0	C-10 H-18 O	**4**_286;	Aetheroleum **R**osmarini
			4_297	Folium **R**osmarini
bornyl acetate	20347-65-3	C-12 H-20 O-2	**1**_271	Radix **V**alerianae
bornyl isovalerate	76-50-6	C-15 H-26 O-2	**1**_271	Radix **V**alerianae
α-boswellic acid	471-66-9	C-30 H-48 O-3	**4**_51	Gummi **B**oswellii
α-boswellic acid acetate (see 3-*O*-acetyl-α-boswellic acid)				
β-boswellic acid	631-69-6	C-30 H-48 O-3	**4**_51	Gummi **B**oswellii
β-boswellic acid acetate (see 3-*O*-acetyl-β-boswellic acid)				
bruceantin	41451-75-6	C-28 H-36 O-11	**1**_62	Fructus **B**ruceae
brucein A	25514-31-2	C-26 H-34 O-11	**1**_62	Fructus **B**ruceae
brucein B	25514-29-8	C-23 H-28 O-11	**1**_62	Fructus **B**ruceae
brucein C	25514-30-1	C-28 H-36 O-13	**1**_62	Fructus **B**ruceae
brucein D	21499-66-1	C-20 H-26 O-9	**1**_62	Fructus **B**ruceae
brusatol	14907-98-3	C-26 H-32 O-12	**1**_62	Fructus **B**ruceae
buddlejoside A (see agnuside)				
(Z)-butylidenephtalide	551-08-6	C-12 H-12 O-2	**2**_28	Radix **A**ngelicae Sinensis
β-cadinene	523-47-7	C-15 H-24	**2**_28	Radix **A**ngelicae Sinensis
caffeic acid	331-39-5	C-9 H-8 O-4	**4**_95	Folium **C**ynarae
3-caffeoylquinic acid (see chlorogenic acid)				
cajeputol (see 1,8-cineole)				
calenduloside A	32725-74-9	C-42 H-68 O-13	**2**_38	Flos **C**alendulae
calenduloside E	26020-14-4	C-36 H-56 O-9	**2**_38	Flos **C**alendulae
calenduloside F	51415-02-2	C-42 H-66 O-14	**2**_38	Flos **C**alendulae
calenduloside H	26020-29-1	C-48 H-76 O-19	**2**_38	Flos **C**alendulae
camphol (see borneol)				
camphor	76-22-2	C-10 H-16 O	**4**_184;	Herba **M**illefolii

Name	CAS RN	Mol. form.	Volume	Monograph name
			4_286;	Aetheroleum **R**osmarini
			4_297	Folium **R**osmarini
dl-camphor (see camphor)				
(+)-(*R*)-canadine (see β-canadine)				
(-)-(*S*)-canadine (see α-canadine)				
α-canadine	5096-57-1	C-20 H-21 N O-4	**3**_197	Rhizoma **H**ydrastis
β-canadine	2086-96-6	C-20 H-21 N O-4	**3**_197	Rhizoma **H**ydrastis
canin	24959-84-0	C-15 H-18 O-5	**2**_320	Herba **T**anaceti Parthenii
cannabiscetin (see myricetin)				
carnosol	5957-80-2	C-20 H-26 O-4	**4**_297	Folium **R**osmarini
carthamin	36338-96-2	C-43 H-42 O-22	**3**_117	Flos **C**arthami
carvacrol	499-75-2	C-10 H-14 O	**1**_262;	Herba **T**hymi
			2_28	Radix **A**ngelicae Sinensis
carvone	99-49-0	C-10 H-14 O	**3**_36	Fructus **A**nethi
caryophyllene (see β-caryophyllene)				
α-caryophyllene (see humulene)				
β-caryophyllene	87-44-5	C-15 H-24	**2**_48;	Flos **C**aryophylli
			2_183;	Folium **M**elissae
			2_209;	Folium **O**cimi Sancti
			4_130;	Folium **G**uavae
			4_184	Herba **M**illefolii
cascaroside A	53823-08-8	C-27 H-32 O-14	**2**_262	Cortex **R**hamni Purshianae
cascaroside B	53861-34-0	C-27 H-32 O-14	**2**_262	Cortex **R**hamni Purshianae
cascaroside C	53823-09-9	C-27 H-32 O-13	**2**_262	Cortex **R**hamni Purshianae
cascaroside D	53861-35-1	C-27 H-32 O-13	**2**_262	Cortex **R**hamni Purshianae
cascaroside E	164178-32-9	C-27 H-32 O-14	**2**_262	Cortex **R**hamni Purshianae
cascaroside F	164322-83-2	C-27 H-32 O-14	**2**_262	Cortex **R**hamni Purshianae
casticin	479-91-4	C-19 H-18 O-8	**4**_13	Fructus **A**gni Casti
catalpol	2415-24-9	C-15 H-22 O-10	**3**_286	Radix **R**ehmanniae
catechin	154-23-4	C-15 H-14 O-6	**2**_70	Folium cum Flore **C**rataegi
catechol (see catechin)				
cathine (see (+)-norpseudoephedrine)				
cembrene	1898-13-1	C-20 H-32	**3**_172	Gummi **G**ugguli
cepaenes			**1**_8	Bulbus **A**llii Cepae
cephaeline	483-17-0	C-28 H-38 N-2 O-4	**3**_208	Radix **I**pecacuanhae

Name	CAS RN	Mol. form.	Volume	Monograph name
ceramides	104404-17-3		**2**_333	Radix **U**rticae
cetraric acid	489-49-6	C-20 H-18 O-9	**4**_143	Lichen **I**slandicus
chamazulene	529-05-5	C-14 H-16	**1**_89;	Flos **C**hamomillae
			4_184	Herba **M**illefolii
chebulagic acid	23094-71-5	C-41 H-30 O-27	**4**_74	Fructus **C**hebulae
chebulanin	166833-80-3	C-27 H-24 O-19	**4**_74	Fructus **C**hebulae
chebulinic acid	18942-26-2	C-41 H-32 O-27	**4**_74	Fructus **C**hebulae
chicoric acid	6537-80-0	C-22 H-18 O-12	**1**_130;	Radix **E**chinaceae
			1_139	Herba **E**chinaceae Purpureae
chlorogenic acid	327-97-9	C-16 H-18 O-9	**2**_272;	Flos **S**ambuci
			4_95	Folium **C**ynarae
chrysaloin	73889-54-0	C-21 H-22 O-8	**2**_262	Cortex **R**hamni Purshianae
chrysaloin A (see 11-deoxyaloin)				
chrysaloin B (see 11-deoxyaloin)				
chrysanthemin (see cyanidin 3-monoglucoside)				
chrysophanol	481-74-3	C-15 H-10 O-4	**1**_234	Rhizoma **R**hei
chrysosplenol D	14965-20-9	C-18 H-16 O-8	**4**_13	Fructus **A**gni Casti
cichorigenin (see esculetin)				
cimicifugoside	66176-93-0	C-37 H-54 O-11	**2**_58	Rhizoma **C**imicifugae Racemosae
cimigenol	3779-59-7	C-30 H-48 O-5	**2**_58	Rhizoma **C**imicifugae Racemosae
cineole (see 1,8-cineole)				
1,8-cineole	470-82-6	C-10 H-18 O	**2**_99;	Aetheroleum **E**ucalypti
			2_109;	Folium **E**ucalypti
			2_174;	Aetheroleum **M**elaleucae Alternifoliae
			3_36;	Fructus **A**nethi
			4_69;	Semen **C**ardamomi
			4_184;	Herba **M**illefolii
			4_286;	Aetheroleum **R**osmarini
			4_297	Folium **R**osmarini
cinnamaldehyde	104-55-2	C-9 H-8 O	**1**_99	Cortex **C**innamomi
6´-cinnamoylcatalpol (see picroside I)				
citral A (see geranial)				
citral b (see neral)				
citronellal	106-23-0	C-10 H-18 O	**2**_183	Folium **M**elissae
columbamine	3621-36-1	C-20 H-20 N O-4	**4**_33	Cortex **B**erberidis
coniferyl ferulate	63644-62-2	C-20 H-20 O-6	**2**_28	Radix **A**ngelicae Sinensis

Name	CAS RN	Mol. form.	Volume	Monograph name
corynoxeine	630-94-4	C-22 H-26 N-2 O-4	**4**_356	Ramulus cum Uncis **U**ncariae
coumarin	91-64-5	C-9 H-6 O-2	**1**_99	Cortex **C**innamomi
crocetin (see α-crocetin)				
α-crocetin	27876-94-4	C-20 H-24 O-4	**3**_129	Stigma **C**roci
β-crocetin	25368-09-6	C-21 H-26 O-4	**3**_129	Stigma **C**roci
γ-crocetin	5892-54-6	C-22 H-28 O-4	**3**_129	Stigma **C**roci
crocin (see A-crocin)				
A-crocin	42553-65-1	C-44 H-64 O-24	**3**_129	Stigma **C**roci
B-crocin	55750-84-0	C-38 H-54 O-19	**3**_129	Stigma **C**roci
C-crocin	55750-85-1	C-32 H-44 O-14	**3**_129	Stigma **C**roci
D-crocin	57710-64-2	C-32 H-44 O-14	**3**_129	Stigma **C**roci
E-crocin	58050-17-2	C-26 H-34 O-9	**3**_129	Stigma **C**roci
crocin 2 (see B-crocin)				
crocin 3 (see C-crocin)				
crocin 4 (see D-crocin)				
cucurbitacin B	6199-67-3	C-32 H-46 O-8	**4**_262	Rhizoma **P**icrorhizae
cucurbitacin D	3877-86-9	C-30 H-44 O-7	**4**_262	Rhizoma **P**icrorhizae
cucurbitacin E	18444-66-1	C-32 H-44 O-8	**4**_262	Rhizoma **P**icrorhizae
cucurbitacin F	5939-57-1	C-30 H-46 O-7	**4**_262	Rhizoma **P**icrorhizae
cucurbitacin I	2222-07-3	C-30 H-42 O-7	**4**_262	Rhizoma **P**icrorhizae
cucurbitacin R	55903-92-9	C-30 H-46 O-7	**4**_262	Rhizoma **P**icrorhizae
cucurbitine	6807-92-7	C-5 H-10 N-2 O-2	**4**_86	Semen **C**ucurbitae
curcumin	458-37-7	C-21 H-20 O-6	**1**_118	Rhizoma **C**urcumae Longae
curcumin I (see curcumin)				
curcumin II (see desmethoxycurcumin)				
curcumin III (see bisdesmethoxycurcumin)				
curzerene	17910-09-7	C-15 H-20 O	**3**_250	Gummi **M**yrrha
curzerenone	20493-56-5	C-15 H-18 O-2	**3**_250	Gummi **M**yrrha
cuscohygrine	454-14-8	C-13 H-24 N-2 O	**4**_376	Radix **W**ithaniae
cyanidin	528-58-5	C-15 H-11 Cl O-6	**4**_152	Fructus **M**acrocarponii
cyanidin 3-monoglucoside	7084-24-4	C-21 H-21 Cl O-11	**4**_213	Fructus **M**yrtilli
cyanidol (see cyanidin)				
cyclamin (see malvidin 3-monoglucoside)				
p-cymene	99-87-6	C-10 H-14	**3**_36	Fructus **A**nethi
cynarin	30964-13-7	C-25 H-24 O-12	**1**_130;	Radix **E**chinaceae
			4_95	Folium **C**ynarae
cynaropicrin	35730-78-0	C-19 H-22 O-6	**4**_95	Folium **C**ynarae
cynaroside	5373-11-5	C-21 H-20 O-11	**4**_13;	Fructus **A**gni Casti
			4_95	Folium **C**ynarae
daidzein	486-66-8	C-15 H-10 O-4	**4**_338	Flos **T**rifolii

Name	CAS RN	Mol. form.	Volume	Monograph name
daidzeol (see daidzein)				
daucosterol	474-58-8	C-35 H-60 O-6	**2**_86;	Radix **E**leutherococci
			2_333	Radix **U**rticae
deacetylnimbin	18609-16-0	C-28 H-34 O-8	**3**_91;	Folium **A**zadirachti
			3_104	Oleum **A**zadirachti
delphinidin 3-monoglucoside	6906-38-3	C-21 H-21 Cl O-12	**4**_213	Fructus **M**yrtilli
delphinin (see delphinidin 3-monoglucoside)				
demethoxyyangonin	15345-89-8	C-14 H-12 O-3	**2**_234	Rhizoma **P**iperis Methystici
(+)-11-deoxyaloin	69980-49-0	C-21 H-22 O-8	**2**_262	Cortex **R**hamni Purshianae
(-)-11-deoxyaloin	18262-45-8	C-21 H-22 O-8	**2**_262	Cortex **R**hamni Purshianae
deoxyschizandrin; wuweizisu A (see schisandrin A)				
desacylsenegasaponin A	173557-02-3	C-64 H-102 O-33	**2**_279	Radix **S**enegae
desacylsenegin II	163589-51-3	C-59 H-94 O-29	**2**_279	Radix **S**enegae
desacylsenegin III	180572-65-0	C-65 H-104 O-33	**2**_279	Radix **S**enegae
desmethoxycurcumin	22608-11-3	C-20 H-18 O-5	**1**_118	Rhizoma **C**urcumae Longae
diallyl disulfide	2179-57-9	C-6 H-10 S-2	**1**_19	Bulbus **A**lli Sativi
diallyl trisulfide	2050-87-5	C-6 H-10 S-3	**1**_19	Bulbus **A**lli Sativi
1,5-dicaffeoylquinic acid (see cynarin)				
1,5-dicaffeylquinic acid (see cynarin)				
dihydrocarvone	5948-04-9	C-10 H-16 O	**3**_36	Fructus **A**nethi
(9*E*)-4,5-dihydro-furanodien-6-one	88010-65-5	C-15 H-20 O-2	**3**_250	Gummi **M**yrrha
11α,13-dihydro-helenalin	34257-95-9	C-15 H-20 O-4	**3**_80	Flos **A**rnicae
dihydrokawain	587-63-3	C-14 H-16 O-3	**2**_234	Rhizoma **P**iperis Methystici
dihydromethysticin	19902-91-1	C-15 H-16 O-5	**2**_234	Rhizoma **P**iperis Methystici
3,4-dihydroxycinnamic acid (see caffeic acid)				
dimethulene (see chamazulene)				
dimethylcrocetin (see γ-crocetin)				
α-dimorphecolic acid	73543-67-6	C-18 H-32 O-3	**2**_333	Radix **U**rticae
docosanol	661-19-8	C-22 H-46 O	**2**_248	Cortex **P**runi Africanae
1-docosanol (see docosanol)				
docosyl (*E*)-ferulate	101927-24-6	C-32 H-54 O-4	**2**_248	Cortex **P**runi Africanae
echinacoside	82854-37-3	C-35 H-46 O-20	**1**_130	Radix **E**chinaceae
(+)-echinolone	59440-97-0	C-14 H-24 O-2	**1**_130	Radix **E**chinaceae
elatericin B (see cucurbitacin I)				

Name	CAS RN	Mol. form.	Volume	Monograph name
elatericine A (see cucurbitacin D)				
α-elaterine (see cucurbitacin E)				
eleutheroside A (see daucosterol)				
eleutheroside B	118-34-3	C-17 H-24 O-9	**2**_86	Radix **E**leutherococci
eleutheroside B1	16845-16-2	C-17 H-20 O-10	**2**_86	Radix **E**leutherococci
eleutheroside B4 (see (+)-sesamin)				
eleutheroside C	15486-24-5	C-8 H-16 O-6	**2**_86	Radix **E**leutherococci
eleutheroside D	79484-75-6	C-34 H-46 O-18	**2**_86	Radix **E**leutherococci
eleutheroside E	39432-56-9	C-34 H-46 O-18	**2**_86	Radix **E**leutherococci
eleutheroside E1	7374-79-0	C-28 H-36 O-13	**2**_86	Radix **E**leutherococci
ellagic acid	476-66-4	C-14 H-6 O-8	**4**_120;	Pericarpium **G**ranati
			4_130	Folium **G**uavae
emetine	483-18-1	C-29 H-40 N-2 O-4	**3**_208	Radix **I**pecacuanhae
emodin (see frangula emodin)				
enin (see malvidin 3-monoglucoside)				
(-)-ephedrine	299-42-3	C-10 H-15 N O	**1**_148	Herba **E**phedrae
epi-catechin	490-46-0	C-15 H-14 O-6	**2**_70	Folium cum Flore **C**rataegi
epi-catechol (*see epi*-catechin)				
eremophilene	10219-75-7	C-15 H-24	**2**_183	Folium **M**elissae
escholine (see magnoflorine)				
esculetin	305-01-1	C-9 H-6 O-4	**4**_369	Cortex **V**iburni Prunifolii
esculetol (see esculetin)				
estragole	140-67-0	C-10 H-12 O	**3**_139	Fructus **F**oeniculi
estragole (see methylchavicol)				
eucalyptol (see 1,8-cineole)				
eugenol	97-53-0	C-10 H-12 O-2	**1**_99;	Cortex **C**innamomi
			2_48;	Flos **C**aryophylli
			2_209	Folium **O**cimi Sancti
eugenol acetate	93-28-7	C-12 H-14 O-3	**2**_48	Flos **C**aryophylli
eutannin (see chebulinic acid)				
farnesyl acetate	4128-17-0	C-17 H-28 O-2	**2**_183	Folium **M**elissae
fenchone	1195-79-5	C-10 H-16 O	**3**_139	Fructus **F**oeniculi
feniculin (see gajaverin)				
(*E*)-ferulic acid	1135-24-6	C-10 H-10 O-4	**2**_28	Radix **A**ngelicae Sinensis
flavosil (see formononetin)				
formononetin	485-72-3	C-16 H-12 O-4	**2**_58;	Rhizoma **C**imicifugae Racemosae
			4_338	Flos **T**rifolii
fragilin	19764-02-4	C-15 H-20 O-8	**4**_312	Cortex **S**alicis
frangula emodin	518-82-1	C-15 H-10 O-5	**1**_234;	Rhizoma **R**hei

Name	CAS RN	Mol. form.	Volume	Monograph name
			2_117	Cortex **F**rangulae
frangula emodin anthrone	491-60-1	C-15 H-12 O-4	**2**_117	Cortex **F**rangulae
frangulin A	521-62-0	C-21 H-20 O-9	**2**_117	Cortex **F**rangulae
frangulin B	14101-04-3	C-20 H-18 O-9	**2**_117	Cortex **F**rangulae
franguloside (see frangulin A)				
friedelin	559-74-0	C-30 H-50 O	**2**_249	Cortex **P**runi Africanae
fumarprotocetraric acid	489-50-9	C-22 H-16 O-12	**4**_143	Lichen **I**slandicus
(5*E*,9*Z*)-furanodiene	19912-61-9	C-15 H-20 O	**3**_250	Gummi **M**yrrha
furanoeudesma-1,3-diene	87605-93-4	C-15 H-18 O	**3**_250	Gummi **M**yrrha
gallic acid	149-91-7	C-7 H-6 O-5	**2**_129;	Folium et Cortex **H**amamelidis
			2_345;	Folium **U**vae Ursi
			4_74	Fructus **C**hebulae
gallogen (see ellagic acid)				
genistein	446-72-0	C-15 H-10 O-5	**4**_338	Flos **T**rifolii
genisteol (see genistein)				
genkwanin	437-64-9	C-16 H-12 O-5	**4**_297	Folium **R**osmarini
gentiopicrin (see gentiopicroside)				
gentiopicroside	20831-76-9	C-16 H-20 O-9	**3**_153;	Radix **G**entianae Luteae
			3_162	Radix **G**entianae Scabrae
geranial	141-27-5	C-10 H-16 O	**2**_183	Folium **M**elissae
geraniol	106-24-1	C-10 H-18 O	**2**_183	Folium **M**elissae
[1]-gingerol	99742-03-7	C-12 H-16 O-4	**1**_280	Rhizoma **Z**ingiberis
[3]-gingerol	41743-67-3	C-14 H-20 O-4	**1**_280	Rhizoma **Z**ingiberis
[4]-gingerol	41743-68-4	C-15 H-22 O-4	**1**_280	Rhizoma **Z**ingiberis
[5]-gingerol	41743-69-5	C-16 H-24 O-4	**1**_280	Rhizoma **Z**ingiberis
[6]-gingerol	23513-14-6	C-17 H-26 O-4	**1**_280	Rhizoma **Z**ingiberis
[8]-gingerol	23513-08-8	C-19 H-30 O-4	**1**_280	Rhizoma **Z**ingiberis
[10]-gingerol	23513-15-7	C-21 H-34 O-4	**1**_280	Rhizoma **Z**ingiberis
ginkgetin	481-46-9	C-32 H-22 O-10	**1**_157	Folium **G**inkgo
ginkgolide A	15291-75-5	C-20 H-24 O-9	**1**_157	Folium **G**inkgo
ginkgolide B	15291-77-7	C-20 H-24 O-10	**1**_157	Folium **G**inkgo
ginkgolide C	15291-76-6	C-20 H-24 O-11	**1**_157	Folium **G**inkgo
ginkgolide J	107438-79-9	C-20 H-24 O-10	**1**_157	Folium **G**inkgo
ginkgolide M	15291-78-8	C-20 H-24 O-10	**1**_157	Folium **G**inkgo
ginsenoside A1 (see pseudoginsenoside F11)				
ginsenoside Rb1	41753-43-9	C-54 H-92 O-23	**1**_171;	Radix **G**inseng
			4_230	Radix **P**anacis Quinquefolii

Name	CAS RN	Mol. form.	Volume	Monograph name
ginsenoside Rb2	11021-13-9	C-53 H-90 O-22	**1**_171;	Radix **G**inseng
			4_230	Radix **P**anacis Quinquefolii
ginsenoside Rc	11021-14-0	C-53 H-90 O-22	**1**_171;	Radix **G**inseng
			4_230	Radix **P**anacis Quinquefolii
ginsenoside Rd	52705-93-8	C-48 H-82 O-18	**1**_171;	Radix **G**inseng
			4_230	Radix **P**anacis Quinquefolii
ginsenoside Re	52286-59-6	C-48 H-82 O-18	**1**_171;	Radix **G**inseng
			4_230	Radix **P**anacis Quinquefolii
ginsenoside Rf	52286-58-5	C-42 H-72 O-14	**1**_171	Radix **G**inseng
ginsenoside Rg1	22427-39-0	C-42 H-72 O-14	**1**_171;	Radix **G**inseng
			4_230	Radix **P**anacis Quinquefolii
ginsenoside Rg2	52286-74-5	C-42 H-72 O-13	**1**_171	Radix **G**inseng
glaucarubinone	1259-86-5	C-25 H-34 O-10	**1**_62	Fructus **B**ruceae
glucofrangulin A	21133-53-9	C-27 H-30 O-14	**2**_117	Cortex **F**rangulae
glucofrangulin B	14062-59-0	C-26 H-28 O-14	**2**_117	Cortex **F**rangulae
glycyrrhetic acid	471-53-4	C-30 H-46 O-4	**1**_187	Radix **G**lycyrrhizae
glycyrrhetinic acid (see glycyrrhetic acid)				
glycyrrhizic acid (see glycyrrhizin)				
glycyrrhizin	1405-86-3	C-42 H-62 O-16	**1**_187	Radix **G**lycyrrhizae
gomisin A (see schisandrol B)				
gomisin N	69176-52-9	C-23 H-28 O-6	**3**_299	Fructus **S**chisandrae
goyaglycoside a	333332-41-5	C-37 H-60 O-9	**4**_195	Fructus **M**omordicae
goyaglycoside b	333332-48-2	C-37 H-60 O-9	**4**_195	Fructus **M**omordicae
goyaglycoside c	333332-49-3	C-38 H-62 O-9	**4**_195	Fructus **M**omordicae
goyaglycoside d	333332-50-6	C-38 H-62 O-9	**4**_195	Fructus **M**omordicae
goyaglycoside e	333333-12-3	C-42 H-68 O-13	**4**_195	Fructus **M**omordicae
goyaglycoside f	333333-13-4	C-42 H-68 O-13	**4**_195	Fructus **M**omordicae
goyaglycoside g	333333-14-5	C-43 H-70 O-14	**4**_195	Fructus **M**omordicae
goyaglycoside h	333333-19-0	C-42 H-70 O-15	**4**_195	Fructus **M**omordicae
goyasaponin I	333333-20-3	C-65 H-102 O-31	**4**_195	Fructus **M**omordicae
goyasaponin II	333333-23-6	C-70 H-110 O-35	**4**_195	Fructus **M**omordicae
goyasaponin III	333333-27-0	C-49 H-76 O-19	**4**_195	Fructus **M**omordicae
guajaverin	22255-13-6	C-20 H-18 O-11	**4**_130	Folium **G**uavae
guggulsterol I	39025-25-7	C-27 H-44 O-4	**3**_172	Gummi **G**ugguli
guggulsterol-II	39025-26-8	C-27 H-46 O-3	**3**_172	Gummi **G**ugguli
guggulsterol-III	39025-27-9	C-27 H-44 O-3	**3**_172	Gummi **G**ugguli
(*E*)-guggulsterone	39025-24-6	C-21 H-28 O-2	**3**_172	Gummi **G**ugguli
(*Z*)-guggulsterone	39025-23-5	C-21 H-28 O-2	**3**_172	Gummi **G**ugguli

Name	CAS RN	Mol. form.	Volume	Monograph name
gypenoside VIII (see ginsenoside Rd)				
hamamelitannin	469-32-9	C-20 H-20 O-14	**2**_129	Folium et Cortex **H**amamelidis
harman	486-84-0	C-12 H-10 N-2	**3**_261	Herba **P**assiflorae
harpagide	6926-08-5	C-15 H-24 O-10	**3**_185	Radix **H**arpagophyti
harpagoside	19210-12-9	C-24 H-30 O-11	**3**_185	Radix **H**arpagophyti
helenalin	6754-13-8	C-15 H-18 O-4	**3**_80	Flos **A**rnicae
henquanin (see genkwanin)				
heraclin (see bergapten)				
homobaldrinal	67910-07-0	C-15 H-16 O-4	**1**_271	Radix **V**alerianae
homovanillic alcohol (see homovanillyl alcohol)				
homovanillyl alcohol	2380-78-1	C-9 H-12 O-3	**2**_333	Radix **U**rticae
honokiol	35354-74-6	C-18 H-18 O-2	**4**_170	Cortex **M**agnoliae
α-humulene (see humulene)				
humulene (α-caryophyllene)	6753-98-6	C-15 H-24	**2**_48;	Flos **C**aryophylli
			2_183;	Folium **M**elissae
			2_209;	Folium **O**cimi Sancti
			3_239	Strobilus **L**upuli
humulone	26472-41-3	C-21 H-30 O-5	**3**_239	Strobilus **L**upuli
hydrastine	118-08-1	C-21 H-21 N O-6	**3**_197	Rhizoma **H**ydrastis
hydroquinone	123-31-9	C-6 H-6 O-2	**2**_345	Folium **U**vae Ursi
p-hydroxybenzaldehyde	123-08-0	C-7 H-6 O-2	**2**_333	Radix **U**rticae
3β-hydroxyparthenolide		C-15 H-20 O-4	**2**_320	Herba **T**anaceti Parthenii
hydroxysafflor yellow A	78281-02-4	C-27 H-32 O-16	**3**_117	Flos **C**arthami
hyperforin	11079-53-1	C-35 H-52 O-4	**2**_153	Herba **H**yperici
hypericin	548-04-9	C-30 H-16 O-8	**2**_153	Herba **H**yperici
hyperoside	482-36-0	C-21 H-20 O-12	**2**_39;	Flos **C**alendulae
			2_70;	Folium cum Flore **C**rataegi
			2_153	Herba **H**yperici
imperatorin	482-44-0	C-16 H-14 O-4	**3**_12	Fructus **A**mmi Majoris
ipecoside	15401-60-2	C-27 H-35 N O-12	**3**_208	Radix **I**pecacuanhae
isoanethole (see methylchavicol)				
isoastragaloside I	84676-88-0	C-45 H-72 O-16	**1**_53	Radix **A**stragali
isoastragaloside II	86764-11-6	C-43 H-70 O-15	**1**_53	Radix **A**stragali
(10*E*)-*N*-isobutyldodecatetraenamide	75917-90-7	C-16 H-25 N O	**1**_130	Radix **E**chinaceae
(10*Z*)-*N*-isobutyldodecatetraenamide	77448-63-6	C-16 H-25 N O	**1**_130	Radix **E**chinaceae

Name	CAS RN	Mol. form.	Volume	Monograph name
N-isobutyl-dodecatetraenamide	360762-35-2	C-16 H-25 N O	**1**_130	Radix **E**chinaceae
isocorynoxeine	51014-29-0	C-22 H-26 N-2 O-4	**4**_356	Ramulus cum Uncis **U**ncariae
7-isocorynoxeine (see isocorynoxeine)				
(*E*)-isoferulic acid	537-73-5	C-10 H-10 O-4	**2**_58	Rhizoma **C**imicifugae Racemosae
isofraxidin	486-21-5	C-11 H-10 O-5	**2**_86	Radix **E**leutherococci
isoginkgetin	548-19-6	C-32 H-22 O-10	**1**_157	Folium **G**inkgo
isoliquiritigenin	961-29-5	C-15 H-12 O-4	**1**_187	Radix **G**lycyrrhizae
isoliquiritin	5041-81-6	C-21 H-22 O-9	**1**_187	Radix **G**lycyrrhizae
isomitraphylline	4963-01-3	C-21 H-24 N-2 O-4	**3**_352	Cortex **U**ncariae
iso-orientin	4261-42-1	C-21 H-20 O-11	**3**_261	Herba **P**assiflorae
isopelletierine	4396-01-4	C-8 H-15 N O	**4**_376	Radix **W**ithaniae
isopteropodine	5171-37-9	C-21 H-24 N-2 O-4	**3**_352	Cortex **U**ncariae
isopunicine (see isopelletierine)				
isoquercitrin	21637-25-2	C-21 H-20 O-12	**2**_39;	Flos **C**alendulae
			2_153;	Herba **H**yperici
			2_272	Flos **S**ambuci
isorhamnetin	480-19-3	C-16 H-12 O-7	**1**_157	Folium **G**inkgo
isorhynchophylline	6859-01-4	C-22 H-28 N-2 O-4	**4**_356	Ramulus cum Uncis **U**ncariae
isoschaftoside	52012-29-0	C-26 H-28 O-14	**3**_261	Herba **P**assiflorae
isotussilagine	91108-32-6	C-10 H-17 N O-3	**1**_130;	Radix **E**chinaceae
			1_139	Herba **E**chinaceae Purpureae
isovaltrate	31078-10-1	C-22 H-30 O-8	**1**_271	Radix **V**alerianae
isovitexin	38953-85-4	C-21 H-20 O-10	**3**_261	Herba **P**assiflorae
jatrorrhizine	3621-38-3	C-20 H-20 N O-4	**4**_33;	Cortex **B**erberidis
			4_247	Cortex **P**hellodendron
jujuboside B	55466-05-2	C-52 H-84 O-21	**3**_362	Fructus **Z**izyphi
kaempferol diglycoside coumarate	111957-48-3	C-36 H-36 O-17	**1**_157	Folium **G**inkgo
kawain	500-64-1	C-14 H-14 O-3	**2**_234	Rhizoma **P**iperis Methystici
11-keto-β-boswellic acid (see 11-oxo-β-boswellic acid)				
11-keto-β-boswellic acid acetate (see 3-*O*-acetyl-11-oxo-β-boswellic acid)				
khellenin (see khellinin)				
khellin	82-02-0	C-14 H-12 O-5	**3**_26	Fructus **A**mmi Visnagae
khellinin	17226-75-4	C-19 H-20 O-10	**3**_26	Fructus **A**mmi Visnagae
khellinol	478-42-2	C-13 H-10 O-5	**3**_26	Fructus **A**mmi Visnagae
khellol	478-79-5	C-13 H-10 O-5	**3**_26	Fructus **A**mmi Visnagae

Name	CAS RN	Mol. form.	Volume	Monograph name
kuromanin (see cyanidin 3-monoglucoside)				
kutkoside	35988-27-3	C-23 H-28 O-13	**4**_262	Rhizoma **P**icrorhizae
lagistase (see ellagic acid)				
lauric acid	143-07-7	C-12 H-24 O-2	**2**_288	Fructus **S**erenoae Repentis
(Z)-ligusticum lactone (see (Z)-butylidenephtalide)				
ligusticumic acid (see valerophenone-*o*-carboxylic acid)				
(Z)-ligustilide	4431-01-0	C-12 H-14 O-2	**2**_28	Radix **A**ngelicae Sinensis
(+)-limonene	5989-27-5	C-10 H-16	**3**_36;	Fructus **A**nethi
			3_139	Fructus **F**oeniculi
linalool (β-linalool)	78-70-6	C-10 H-18 O	**2**_183;	Folium **M**elissae
			3_44;	Aetheroleum **A**nisi
			3_56;	Fructus **A**nisi
			3_221;	Aetheroleum **L**avandulae
			3_232;	Flos **L**avandulae
			4_184	Herba **M**illefolii
linalyl acetate	115-95-7	C-12 H-20 O-2	**3**_221;	Aetheroleum **L**avandulae
			3_232	Flos **L**avandulae
linoleic acid	60-33-3	C-18 H-32 O-2	**2**_219;	Oleum **O**enotherae Biennis
			2_248;	Cortex **P**runi Africanae
			2_288	Fructus **S**erenoae Repentis
cis-linoleic acid (see linoleic acid)				
linolenic acid	463-40-1	C-18 H-30 O-2	**2**_288	Fructus **S**erenoae Repentis
cis-γ-linolenic acid (see γ-linolenic acid)				
γ-linolenic acid	506-26-3	C-18 H-30 O-2	**2**_219	Oleum **O**enotherae Biennis
liquiritigenin	578-86-9	C-15 H-12 O-4	**1**_187	Radix **G**lycyrrhizae
liquiritin	551-15-5	C-21 H-22 O-9	**1**_187	Radix **G**lycyrrhizae
lucenin-2	29428-58-8	C-27 H-30 O-16	**3**_261	Herba **P**assiflorae
lupulone	468-28-0	C-26 H-38 O-4	**3**_239	Strobilus **L**upuli
luteolin 7-*O*-β-D-glucopyranoside (see cynaroside)				
luteolin 7-*O*-β-D-rutinoside (see scolymoside)				
lutexin (see orientin)				
lutonaretin (see iso-orientin)				
madecassic acid	18449-41-7	C-30 H-48 O-6	**1**_80	Herba **C**entellae
madecassoside	34540-22-2	C-48 H-78 O-20	**1**_80	Herba **C**entellae
magnocurarine	6801-40-7	C-19 H-24 N O-3	**4**_170	Cortex **M**agnoliae
magnoflorine	2141-09-5	C-20 H-24 N O-4	**4**_33;	Cortex **B**erberidis

Name	CAS RN	Mol. form.	Volume	Monograph name
			4_247	Cortex **P**hellodendron
magnolol	528-43-8	C-18 H-18 O-2	**4**_170	Cortex **M**agnoliae
majarine (see berberine)				
majudin (see bergapten)				
maltol	118-71-8	C-6 H-6 O-3	**3**_261	Herba **P**assiflorae
malvidin 3-mono-glucoside	7228-78-6	C-23 H-25 Cl O-12	**4**_213	Fructus **M**yrtilli
mamorekku RUH 2 (see rosmarinic acid)				
maslinic acid	4373-41-5	C-30 H-48 O-4	**3**_362	Fructus **Z**izyphi
melittoside	19467-03-9	C-21 H-32 O-15	**3**_286	Radix **R**ehmanniae
menthol	2216-51-5	C-10 H-20 O	**2**_190;	Aetheroleum **M**enthae Piperitae
			2_202	Folium **M**enthae Piperitae
menthone	14073-97-3	C-10 H-18 O	**2**_190;	Aetheroleum **M**enthae Piperitae
			2_202	Folium **M**enthae Piperitae
methoxsalen (see xanthotoxin)				
5´-methoxybilobetin	77053-35-1	C-32 H-22 O-11	**1**_157	Folium **G**inkgo
(5*E*,8*R*,9*E*)-2-methox-yfuranodiene	108376-98-3	C-16 H-22 O-2	**3**_250	Gummi **M**yrrha
(5*E*,8*S*,9*E*)-2-methox-yfuranodiene	82901-10-8	C-16 H-22 O-2	**3**_250	Gummi **M**yrrha
5-methoxypsoralen (see bergapten)				
8-methoxypsoralen (see xanthotoxin)				
7-methylaxillarin (see chrysosplenol D)				
2-methylbut-3-en-2-ol	115-18-4	C-5 H-10 O	**3**_239	Strobilus **L**upuli
methylchavicol	140-67-0	C-10 H-12 O	**3**_44;	Aetheroleum **A**nisi
			3_56	Fructus **A**nisi
N-methylcoreximine (see phellodendrine)				
methylcrocetin (see β-crocetin)				
methylcysteine *S*-oxide	6853-87-8	C-4 H-9 N O-3 S	**1**_8	Bulbus **A**llii Cepae
(-)-methylephedrine	552-79-4	C-11 H-17 N O	**1**_148	Herba **E**phedrae
methyleugenol	93-15-2	C-11 H-14 O-2	**2**_209	Folium **O**cimi Sancti
N-methyliso-pelletierine	18747-42-7	C-9 H-17 N O	**4**_111	Cortex **G**ranati
methylisopunicine (see *N*-methylisopelletierine)				
3´-methylmyricetin	53472-37-0	C-16 H-12 O-8	**1**_157	Folium **G**inkgo
(+)-methylpseu-doephedrine	51018-28-1	C-11 H-17 N O	**1**_148	Herba **E**phedrae
O-methylpsychotrine	523-01-3	C-29 H-38 N-2 O-4	**3**_208	Radix **I**pecacuanhae
methysticin	495-85-2	C-15 H-14 O-5	**2**_234	Rhizoma **P**iperis Methystici

Name	CAS RN	Mol. form.	Volume	Monograph name
mitraphylline	509-80-8	C-21 H-24 N-2 O-4	**3**_352	Cortex **U**ncariae
mitrinermine (see rhynchophylline)				
momordicoside F1	81348-81-4	C-37 H-60 O-8	**4**_195	Fructus **M**omordicae
momordicoside F2	81348-82-5	C-36 H-58 O-8	**4**_195	Fructus **M**omordicae
momordicoside G	81371-54-2	C-37 H-60 O-8	**4**_195	Fructus **M**omordicae
momordicoside I	81371-55-3	C-36 H-58 O-8	**4**_195	Fructus **M**omordicae
momordicoside K	81348-84-7	C-37 H-60 O-9	**4**_195	Fructus **M**omordicae
momordicoside L	81348-83-6	C-36 H-58 O-9	**4**_195	Fructus **M**omordicae
monomelittoside	20633-72-1	C-15 H-22 O-10	**3**_286	Radix **R**ehmanniae
mukulol	41943-03-7	C-20 H-34 O	**3**_172	Gummi **G**ugguli
myricetin	529-44-2	C-15 H-10 O-8	**1**_157;	Folium **G**inkgo
			4_152	Fructus **M**acrocarponii
myricetol (see myricetin)				
myrtillin (see delphinidin 3-monoglucoside)				
neoandrographolide	27215-14-1	C-26 H-40 O-8	**2**_15	Herba **A**ndrographidis
neocarthamin	519-54-0	C-21 H-22 O-11	**3**_117	Flos **C**arthami
neoolivil	77790-55-7	C-20 H-24 O-7	**2**_333	Radix **U**rticae
neprotine (see jatrorrhizine)				
neral	106-26-3	C-10 H-16 O	**2**_183	Folium **M**elissae
nerol	106-25-2	C-10 H-18 O	**2**_183	Folium **M**elissae
nimbin	5945-86-8	C-30 H-36 O-9	**3**_91;	Folium **A**zadirachti
			3_104	Oleum **A**zadirachti
nopinene (see β-pinene)				
(-)-norephedrine	492-41-1	C-9 H-13 N O	**1**_148	Herba **E**phedrae
(+)-norpseudoephedrine	492-39-7	C-9 H-13 N O	**1**_148	Herba **E**phedrae
cis-β-ocimene	3338-55-4	C-10 H-16	**2**_28	Radix **A**ngelicae Sinensis
oleanolic acid	508-02-1	C-30 H-48 O-3	**2**_333;	Radix **U**rticae
			4_130	Folium **G**uavae
oleic acid	112-80-1	C-18 H-34 O-2	**2**_248	Cortex **P**runi Africanae
olmelin (see biochanin A)				
orientin	28608-75-5	C-21 H-20 O-11	**3**_261	Herba **P**assiflorae
11-oxo-β-boswellic acid	17019-92-0	C-30 H-46 O-4	**4**_51	Gummi **B**oswellii
paeoniflorin	23180-57-6	C-23 H-28 O-11	**1**_197	Radix **P**aeoniae
palmatine	3486-67-7	C-21 H-22 N O-4	**4**_247	Cortex **P**hellodendron
panaxadiol	19666-76-3	C-30 H-52 O-3	**1**_171	Radix **G**inseng
panaxatriol	32791-84-7	C-30 H-52 O-4	**1**_171	Radix **G**inseng
panaxoside A (see ginsenoside Rg1)				
panaxoside Rb1 (see ginsenoside Rb1)				

Name	CAS RN	Mol. form.	Volume	Monograph name
panaxoside Rc (see ginsenoside Rc)				
panaxoside Re (see ginsenoside Re)				
parthenolide	20554-84-1	C-15 H-20 O-3	**2**_320	Herba **T**anaceti Parthenii
patrinoside	53962-20-2	C-21 H-34 O-11	**4**_369	Cortex **V**iburni Prunifolii
pelletierine	2858-66-4	C-8 H-15 N O	**4**_111	Cortex **G**ranati
peonidin	134-01-0	C-16 H-13 Cl O-6	**4**_152	Fructus **M**acrocarponii
peonidin 3-mono-glucoside	6906-39-4	C-22 H-23 Cl O-11	**4**_213	Fructus **M**yrtilli
peonidol chloride (see peonidin)				
petunidin 3-mono-glucoside	6988-81-4	C-22 H-23 Cl O-12	**4**_213	Fructus **M**yrtilli
(-)-α-phellandrene	4221-98-1	C-10 H-16	**3**_36;	Fructus **A**nethi
			3_139	Fructus **F**oeniculi
phellodendrine	6873-13-8	C-20 H-24 N O-4	**4**_247	Cortex **P**hellodendron
physcione	521-61-9	C-16 H-12 O-5	**1**_234	Rhizoma **R**hei
picrocrocin	138-55-6	C-16 H-26 O-7	**3**_129	Stigma **C**roci
picrosalvin (see carnosol)				
picroside I	27409-30-9	C-24 H-28 O-11	**4**_262	Rhizoma **P**icrorhizae
picroside II	39012-20-9	C-23 H-28 O-13	**4**_262	Rhizoma **P**icrorhizae
picroside III	64461-95-6	C-25 H-30 O-13	**4**_262	Rhizoma **P**icrorhizae
α-pinene	80-56-8	C-10 H-16	**3**_36**;**	Fructus **A**nethi
			3_139;	Fructus **F**oeniculi
			4_130;	Folium **G**uavae
			4_184	Herba **M**illefolii
β-pinene	127-91-3	C-10 H-16	**4**_184	Herba **M**illefolii
platycodin A	66779-34-8	C-59 H-94 O-29	**1**_216	Radix **P**latycodi
platycodin C	66779-35-9	C-59 H-94 O-29	**1**_216	Radix **P**latycodi
platycodin D	58479-68-8	C-57 H-92 O-28	**1**_216	Radix **P**latycodi
platycodin D2	66663-90-9	C-63 H-102 O-33	**1**_216	Radix **P**latycodi
plenolin (see 11α, 13-dihydrohelenalin)				
polygalacin D	66663-91-0	C-57 H-92 O-27	**1**_216	Radix **P**latycodi
polygalacin D2	66663-92-1	C-63 H-102 O-32	**1**_216	Radix **P**latycodi
proanthocyanidin trimer		C-45 H-38 O-18	**4**_152	Fructus **M**acrocarponii
proanthocyanidins		C-30 H-26 O-14	**2**_129	Folium et Cortex **H**amamelidis
procumbide	20486-27-5	C-15 H-22 O-10	**3**_185	Radix **H**arpagophyti
procyanidin	15514-06-4	C-30 H-26 O-12	**2**_129	Folium et Cortex **H**amamelidis
procyanidin B1	20315-25-7	C-30 H-26 O-12	**2**_70	Folium cum Flore **C**rataegi

Name	CAS RN	Mol. form.	Volume	Monograph name
procyanidin B2	29106-49-8	C-30 H-26 O-12	**2**_70	Folium cum Flore **C**rataegi
procyanidin B3	23567-23-9	C-30 H-26 O-12	**2**_70	Folium cum Flore **C**rataegi
procyanidin B4	29106-51-2	C-30 H-26 O-12	**2**_70	Folium cum Flore **C**rataegi
prodelphinidin		C-30 H-26 O-14	**2**_129	Folium et Cortex **H**amamelidis
propenylcysteine oxide		C-6 H-11 N O-3 S	**1**_8	Bulbus **A**llii Cepae
propylcysteine *S*-oxide	17795-24-3	C-6 H-13 N O-3 S	**1**_8	Bulbus **A**llii Cepae
protoaescigenin	20853-07-0	C-30 H-50 O-6	**2**_140	Semen **H**ippocastani
protodioscin	55056-80-9	C-51 H-84 O-22	**4**_326	Fructus **T**ribuli
protoescigenin (see protoaescigenin)				
(+)-protolichesterinic acid	1448-96-0	C-19 H-32 O-4	**4**_143	Lichen **I**slandicus
prunetol (see genistein)				
(+)-pseudoephedrine	90-82-4	C-10 H-15 N O	**1**_148	Herba **E**phedrae
pseudoginsenoside F11	69884-00-0	C-42 H-72 O-14	**4**_230	Radix **P**anacis Quinquefolii
pseudohypericin	55954-61-5	C-30 H-16 O-9	**2**_153	Herba **H**yperici
pseudopelletierine	552-70-5	C-9 H-15 N O	**4**_111	Cortex **G**ranati
pseudopinene (see β-pinene)				
pseudopunicine (see pseudopelletierine)				
pseudo-γ-schisandrin B (see gomisin N)				
pseudotaraxasterol (see ψ-taraxasterol)				
psychotrine	7633-29-6	C-28 H-36 N-2 O-4	**3**_208	Radix **I**pecacuanhae
psyllium	8063-16-9		**1**_208	Semen **P**lantaginis
pteropodine	5629-60-7	C-21 H-24 N-2 O-4	**3**_352	Cortex **U**ncariae
puddumetin (see genkwanin)				
punicalagin	65995-63-3	C-48 H-28 O-30	**4**_111	Cortex **G**ranati
punicalin	65995-64-4	C-34 H-22 O-22	**4**_111;	Cortex **G**ranati
			4_120	Pericarpium **G**ranati
punicine (see pelletierine)				
quassin	76-78-8	C-22 H-28 O-6	**1**_62	Fructus **B**ruceae
quercetin	117-39-5	C-15 H-10 O-7	**2**_272;	Flos **S**ambuci
			4_130	Folium **G**uavae
quercetin diglycoside coumarate	143061-65-8	C-36 H-36 O-18	**1**_157	Folium **G**inkgo
quercitrin	522-12-3	C-21 H-20 O-11	**2**_153	Herba **H**yperici
rehmannioside A	81720-05-0	C-21 H-32 O-15	**3**_286	Radix **R**ehmanniae
rehmannioside B	81720-06-1	C-21 H-32 O-15	**3**_286	Radix **R**ehmanniae
rehmannioside C	81720-07-2	C-21 H-34 O-14	**3**_286	Radix **R**ehmanniae
rehmannioside D	81720-08-3	C-27 H-42 O-20	**3**_286	Radix **R**ehmanniae
rescinnamine	24815-24-5	C-35 H-42 N-2 O-9	**1**_225	Radix **R**auwolfiae

Name	CAS RN	Mol. form.	Volume	Monograph name
reserpine	50-55-5	C-33 H-40 N-2 O-9	**1**_225	Radix **R**auwolfiae
rhein	478-43-3	C-15 H-8 O-6	**1**_234	Rhizoma **R**hei
rheinoside A	111545-28-9	C-27 H-30 O-17	**1**_234	Rhizoma **R**hei
rheinoside B	111614-10-9	C-27 H-30 O-17	**1**_234	Rhizoma **R**hei
rheinoside C	111545-29-0	C-27 H-30 O-16	**1**_234	Rhizoma **R**hei
rheinoside D	111614-11-0	C-27 H-30 O-16	**1**_234	Rhizoma **R**hei
rhimantin (see aucubin)				
rhodinal (see citronellal)				
rhynchophylline	76-66-4	C-22 H-28 N-2 O-4	**4**_356	Ramulus cum Uncis **U**ncariae
Δ-18-rhyncophylline (see corynoxeine)				
ricinic acid (see ricinoleic acid)				
ricinoleic acid	141-22-0	C-18 H-34 O-3	**4**_274	Oleum **R**icini
ricinolic acid (see ricinoleic acid)				
rosemaric acid (see rosmarinic acid)				
rosmarinic acid	20283-92-5	C-18 H-16 O-8	**2**_183;	Folium **M**elissae
			4_297	Folium **R**osmarini
rutin	153-18-4	C-27 H-30 O-16	**2**_39;	Flos **C**alendulae
			2_70;	Folium cum Flore **C**rataegi
			2_153;	Herba **H**yperici
			2_272	Flos **S**ambuci
sabinene	3387-41-5	C-10 H-16	**4**_184	Herba **M**illefolii
safflor yellow A	85532-77-0	C-27 H-30 O-15	**3**_117	Flos **C**arthami
safflor yellow B	91574-92-4	C-48 H-54 O-27	**3**_117	Flos **C**arthami
safranal	116-26-7	C-10 H-14 O	**3**_129	Stigma **C**roci
saikogenin A	5092-09-1	C-30 H-48 O-4	**1**_70	Radix **B**upleuri
saikogenin D	5573-16-0	C-30 H-48 O-4	**1**_70	Radix **B**upleuri
saikogenin F	14356-59-3	C-30 H-48 O-4	**1**_70	Radix **B**upleuri
saikogenin G	18175-79-6	C-30 H-48 O-4	**1**_70	Radix **B**upleuri
saikosaponin A	20736-09-8	C-42 H-68 O-13	**1**_70	Radix **B**upleuri
saikosaponin B1	58558-08-0	C-42 H-68 O-13	**1**_70	Radix **B**upleuri
saikosaponin B2	58316-41-9	C-42 H-68 O-13	**1**_70	Radix **B**upleuri
saikosaponin B3	58316-42-0	C-43 H-72 O-14	**1**_70	Radix **B**upleuri
saikosaponin B4	58558-09-1	C-43 H-72 O-14	**1**_70	Radix **B**upleuri
saikosaponin D	20874-52-6	C-42 H-68 O-13	**1**_70	Radix **B**upleuri
salicin	138-52-3	C-13 H-18 O-7	**4**_312	Cortex **S**alicis
salicin 6´-acetate (see fragilin)				
salicortin	29836-41-7	C-20 H-24 O-10	**4**_312	Cortex **S**alicis
salicoside (see salicin)				
sanchinoside C1 (see ginsenoside Rg1)				
sanchinoside Rb1 (see ginsenoside Rb1)				
sanchinoside Re (see ginsenoside Re)				

Name	CAS RN	Mol. form.	Volume	Monograph name
saponaretin (see isovitexin)				
schaftoside	51938-32-0	C-26 H-28 O-14	**3**_261	Herba **P**assiflorae
schisandrin A	61281-38-7	C-24 H-32 O-6	**3**_299	Fructus **S**chisandrae
schisandrin B	61281-37-6	C-23 H-28 O-6	**3**_299	Fructus **S**chisandrae
schisandrol A	7432-28-2	C-24 H-32 O-7	**3**_299	Fructus **S**chisandrae
schisandrol B	58546-54-6	C-23 H-28 O-7	**3**_299	Fructus **S**chisandrae
schizandrin (see schisandrol A)				
(±)-γ-schizandrin (see schisandrin B)				
sciadopitysin	521-34-6	C-33 H-24 O-10	**1**_157	Folium **G**inkgo
scolymoside	20633-84-5	C-27 H-30 O-15	**4**_95	Folium **C**ynarae
scopoletin	92-61-5	C-10 H-8 O-4	**2**_333	Radix **U**rticae
senecionine	130-01-8	C-18 H-25 N O-5	**1**_130;	Radix **E**chinaceae
			1_139	Herba **E**chinaceae Purpureae
(*E*)-senegasaponin A	162762-97-2	C-74 H-110 O-35	**2**_279	Radix **S**enegae
(*Z*)-senegasaponin A	162613-72-1	C-74 H-110 O-35	**2**_279	Radix **S**enegae
(*E*)-senegasaponin B	162870-58-8	C-69 H-102 O-31	**2**_279	Radix **S**enegae
(*Z*)-senegasaponin B	162613-71-0	C-69 H-102 O-31	**2**_279	Radix **S**enegae
senegin II	34366-31-9	C-70 H-104 O-32	**2**_279	Radix **S**enegae
(*Z*)-senegin II	162681-52-9	C-70 H-104 O-32	**2**_279	Radix **S**enegae
(*E*)-senegin III	35906-36-6	C-75 H-112 O-35	**2**_279	Radix **S**enegae
(*Z*)-senegin III	162681-53-0	C-75 H-112 O-35	**2**_279	Radix **S**enegae
sennoside A	81-27-6	C-42 H-38 O-20	**1**_244;	Folium **S**ennae
			1_253	Fructus **S**ennae
sennoside B	128-57-4	C-42 H-38 O-20	**1**_244;	Folium **S**ennae
			1_253	Fructus **S**ennae
sennoside C	37271-16-2	C-42 H-40 O-19	**1**_244;	Folium **S**ennae
			1_253	Fructus **S**ennae
sennoside D	37271-17-3	C-42 H-40 O-19	**1**_244;	Folium **S**ennae
			1_253	Fructus **S**ennae
sennoside E	11137-63-6	C-44 H-38 O-23	**1**_244;	Folium **S**ennae
			1_253	Fructus **S**ennae
sennoside F	52842-23-6	C-44 H-38 O-23	**1**_244;	Folium **S**ennae
			1_253	Fructus **S**ennae
(+)-sesamin	607-80-7	C-20 H-18 O-6	**2**_86	Radix **E**leutherococci
[6]-shogaol	23513-13-5	C-17 H-24 O-3	**1**_280	Rhizoma **Z**ingiberis
[7]-shogaol	99742-07-1	C-18 H-26 O-3	**1**_280	Rhizoma **Z**ingiberis
[9]-shogaol	99742-08-2	C-20 H-30 O-3	**1**_280	Rhizoma **Z**ingiberis
[11]-shogaol	99742-09-3	C-22 H-34 O-3	**1**_280	Rhizoma **Z**ingiberis
[12]-shogaol	99742-10-6	C-23 H-36 O-3	**1**_280	Rhizoma **Z**ingiberis
silybin	22888-70-6	C-25 H-22 O-10	**2**_303	Fructus **S**ilybi Mariae
silychristin	33889-69-9	C-25 H-22 O-10	**2**_303	Fructus **S**ilybi Mariae

Name	CAS RN	Mol. form.	Volume	Monograph name
silydianin	29782-68-1	C-25 H-22 O-10	**2**_303	Fructus **S**ilybi Mariae
β-sitosterol	83-46-5	C-29 H-50 O	**2**_86**;**	Radix **E**leutherococci
			2_249;	Cortex **P**runi Africanae
			2_333	Radix **U**rticae
sitosterone	1058-61-3	C-29 H-48 O	**2**_249	Cortex **P**runi Africanae
sophoricol (see genistein)				
speciophylline	4697-68-1	C-21 H-24 N-2 O-4	**3**_352	Cortex **U**ncariae
spinacene (see squalene)				
spiraeoside	20229-56-5	C-21 H-2O O-12	**2**_70	Folium cum Flore **C**rataegi
squalene	111-02-4	C-30 H-50	**4**_86	Semen **C**ucurbitae
stictin (see cetraric acid)				
sweroside	14215-86-2	C-16 H-22 O-9	**3**_153;	Radix **G**entianae Luteae
			3_162	Radix **G**entianae Scabrae
sylvapine A (see α-pinene)				
(+)-syringaresinol	21453-69-0	C-22 H-26 O-8	**2**_86	Radix **E**leutherococci
syringenin	20675-96-1	C-11 H-14 O-4	**2**_86	Radix **E**leutherococci
taraxacolide glucoside	75911-12-5	C-21 H-32 O-9	**3**_331	Radix cum Herba **T**araxaci
taraxacoside	98449-40-2	C-18 H-22 O-10	**3**_331	Radix cum Herba **T**araxaci
taraxasterol	1059-14-9	C-30 H-50 O	**3**_331	Radix cum Herba **T**araxaci
γ-taraxasterol	464-98-2	C-30 H-50 O	**3**_331	Radix cum Herba **T**araxaci
taraxinic acid glucoside	75911-14-7	C-21 H-28 O-9	**3**_331	Radix cum Herba **T**araxaci
taxifolin	480-18-2	C-15 H-12 O-7	**2**_303	Fructus **S**ilybi Mariae
terebenthene (see β-pinene)				
terminalic acid (see chebulanin)				
terpan (see 1,8-cineole)				
terpinen-4-ol	562-74-3	C-10 H-18 O	**2**_174	Aetheroleum **M**elaleucae Alternifoliae
α-terpinene	99-86-5	C-10 H-16	**2**_174;	Aetheroleum **M**elaleucae Alternifoliae
			3_36	Fructus **A**nethi
γ-terpinene	99-85-4	C-10 H-16	**2**_174	Aetheroleum **M**elaleucae Alternifoliae
4-terpineol (see terpinen-4-ol)				

Name	CAS RN	Mol. form.	Volume	Monograph name
α-terpineol	98-55-5	C-10 H-18 O	**4**_64	Semen **C**ardamomi
(+)-α-terpinyl acetate	7785-54-8	C-12 H-20 O-2	**4**_64	Semen **C**ardamomi
(*R*)-α-terpinyl acetate (see (+)-α-terpinyl acetate)				
tetracosanol	506-51-4	C-24 H-50 O	**2**_248	Cortex **P**runi Africanae
1-tetracosanol (see tetracosanol)				
tetracosyl (*E*)-ferulate	101927-25-7	C-34 H-58 O-4	**2**_248	Cortex **P**runi Africanae
(3*S*,9*R*)-tetrahydroridentin B	75991-58-1	C-15 H-24 O-4	**3**_331	Radix cum Herba **T**araxaci
thalictrine (see magnoflorine)				
thalsine (see berberine)				
4(10)-thujene (see sabinene)				
thymol	89-83-8	C-10 H-14 O	**1**_262	Herba **T**hymi
tinctormine	149475-43-4	C-27 H-31 N O-14	**3**_117	Flos **C**arthami
tribulosaponin A	311310-49-3	C-51 H-84 O-21	**4**_326	Fructus **T**ribuli
tribulosaponin B	311310-52-8	C-51 H-84 O-22	**4**_326	Fructus **T**ribuli
tribulosin	79974-46-2	C-55 H-90 O-25	**4**_326	Fructus **T**ribuli
tribulusamide A	218622-84-5	C-36 H-36 N-2 O-8	**4**_326	Fructus **T**ribuli
tribulusamide B	218622-86-7	C-36 H-34 N-2 O-9	**4**_326	Fructus **T**ribuli
trigonelline	535-83-1	C-7 H-7 N O-2	**3**_341	Semen **T**rigonellae Foenugraeci
α-turmerone	82508-15-4	C-15 H-22 O	**1**_118	Rhizoma **C**urcumae Longae
ar-turmerone	532-65-0	C-15 H-20 O	**1**_118	Rhizoma **C**urcumae Longae
β-turmerone	82508-14-3	C-15 H-22 O	**1**_118	Rhizoma **C**urcumae Longae
(-)-tussilagine	80151-77-5	C-10 H-17 N O-3	**1**_130; **1**_139	Radix **E**chinaceae Herba **E**chinaceae Purpureae
umbellatin (see berberine)				
umbelliferone	93-35-6	C-9 H-6 O-3	**2**_28	Radix **A**ngelicae Sinensis
uncarine C (see pteropodine)				
uncarine D (see speciophylline)				
uncarine E (see isopteropodine)				
uncarine F	14019-66-0	C-21 H-24 N-2 O-4	**3**_352	Cortex **U**ncariae
uncarinic acid A	206256-62-4	C-40 H-56 O-7	**4**_356	Ramulus cum Uncis **U**ncariae
uncarinic acid B	128748-74-8	C-40 H-56 O-7	**4**_356	Ramulus cum Uncis **U**ncariae

Name	CAS RN	Mol. form.	Volume	Monograph name
ursolic acid	77-52-1	C-30 H-48 O-3	**2**_249;	Cortex **P**runi Africanae
			2_272;	Flos **S**ambuci
			2_345;	Folium **U**vae Ursi
			4_130	Folium **G**uavae
uvaol	545-46-0	C-30 H-50 O-2	**2**_345	Folium **U**vae Ursi
valerenal	4176-16-3	C-15 H-22 O	**1**_271	Radix **V**alerianae
valerenic acid	3569-10-6	C-15 H-22 O-2	**1**_271	Radix **V**alerianae
valerophenone-*o*-carboxylic acid	550-37-8	C-12 H-14 O-3	**2**_28	Radix **A**ngelicae Sinensis
valtrate	18296-44-1	C-22 H-30 O-8	**1**_271	Radix **V**alerianae
valtroxal	71013-41-7	C-17 H-22 O-7	**1**_271	Radix **V**alerianae
vanillic acid	121-34-6	C-8 H-8 O-4	**2**_28	Radix **A**ngelicae Sinensis
6-vanilloylcatalpol (see picroside II)				
vicenin (see vicenin-2)				
vicenin-2	23666-13-9	C-27 H-30 O-15	**3**_261	Herba **P**assiflorae
3-vinyl-1,2-dithiin	62488-53-3	C-6 H-8 S-2	**1**_19	Bulbus **A**lli Sativi
2-vinyl-1,3-dithiin	80028-57-5	C-6 H-8 S-2	**1**_19	Bulbus **A**lli Sativi
visnadin	477-32-7	C-21 H-24 O-7	**3**_26	Fructus **A**mmi Visnagae
visnagin	82-57-5	C-13 H-10 O-4	**3**_26	Fructus **A**mmi Visnagae
vitexicarpin (see casticin)				
vitexilactone	61263-49-8	C-22 H-34 O-5	**4**_13	Fructus **A**gni Casti
vitexin	3681-93-4	C-21 H-20 O-10	**2**_70;	Folium cum Flore **C**rataegi
			3_261	Herba **P**assiflorae
vitexin 2˝-rhamnoside	64820-99-1	C-27 H-30 O-14	**2**_70	Folium cum Flore **C**rataegi
vitexlactam A	459167-05-6	C-22 H-35 N O-4	**4**_13	Fructus **A**gni Casti
withaferin A	5119-48-2	C-28 H-38 O-6	**4**_376	Radix **W**ithaniae
withanolide D	30655-48-2	C-28 H-38 O-6	**4**_376	Radix **W**ithaniae
withasomniferol A	194413-09-7	C-28 H-38 O-7	**4**_376	Radix **W**ithaniae
wogonin	632-85-9	C-16 H-12 O-5	**3**_316	Radix **S**cutellariae
wuweizi alcohol B (see schisandrol B)				
wuweizichun A (see schisandrol A)				
wuweizichun B (see schisandrol B)				
wuweizisu B (see schisandrin B)				
xanthohumol	6754-58-1	C-21 H-22 O-5	**3**_239	Strobilus **L**upuli
xanthotoxin	298-81-7	C-12 H-8 O-4	**3**_12	Fructus **A**mmi Majoris
yangonin	500-62-9	C-15 H-14 O-4	**2**_234	Rhizoma **P**iperis Methystici

Name	CAS RN	Mol. form.	Volume	Monograph name
yatrorizine (see jatrorrhizine)				
zingiberene	495-60-3	C-15 H-24	**1**_118;	Rhizoma **C**urcumae Longae
			1_280	Rhizoma **Z**ingiberis
zizyphus saponin I	77943-56-7	C-47 H-76 O-17	**3**_362	Fructus **Z**izyphi
zizyphus saponin II	77943-83-0	C-47 H-76 O-17	**3**_362	Fructus **Z**izyphi
zizyphus saponin III	77943-54-5	C-52 H-84 O-21	**3**_362	Fructus **Z**izyphi
zwiebelanes	126038-52-6	C-6 H-10 O S-2	**1**_8	Bulbus **A**llii Cepae

Annex 6

Cumulative index of major chemical constituents (*ordered by CAS number*)

Cumulative index of major chemical compounds whose chemical structure drawings were presented in four volumes of WHO monographs on selected medicinal plants

CAS RN	Name
50-55-5	reserpine
60-33-3	linoleic acid
76-22-2	camphor
76-50-6	bornyl isovalerate
76-66-4	rhynchophylline
76-78-8	quassin
77-52-1	ursolic acid
78-70-6	linalool (β-linalool)
80-56-8	α-pinene
81-27-6	sennoside A
82-02-0	khellin
82-57-5	visnagin
83-46-5	β-sitosterol
87-44-5	β-caryophyllene
89-83-8	thymol
90-82-4	(+)-pseudoephedrine
91-64-5	coumarin
92-61-5	scopoletin
93-15-2	methyleugenol
93-28-7	eugenol acetate
93-35-6	umbelliferone
97-53-0	eugenol
98-55-5	α-terpineol
99-49-0	carvone
99-85-4	γ-terpinene
99-86-5	α-terpinene
99-87-6	*p*-cymene
104-55-2	cinnamaldehyde
106-23-0	citronellal
106-24-1	geraniol
106-25-2	nerol
106-26-3	neral
111-02-4	squalene
112-80-1	oleic acid
115-18-4	2-methylbut-3-en-2-ol
115-95-7	linalyl acetate
116-26-7	safranal
117-39-5	quercetin
118-08-1	hydrastine
118-34-3	eleutheroside B
118-71-8	maltol
121-34-6	vanillic acid
123-08-0	*p*-hydroxybenzaldehyde
123-11-5	*p*-anisaldehyde
123-31-9	hydroquinone
127-91-3	β-pinene
128-57-4	sennoside B
130-01-8	senecionine
134-01-0	peonidin
138-52-3	salicin
138-55-6	picrocrocin
140-67-0	estragole
140-67-0	methylchavicol
141-22-0	ricinoleic acid

CAS RN	Name
141-27-5	geranial
143-07-7	lauric acid
149-91-7	gallic acid
153-18-4	rutin
154-23-4	catechin
298-81-7	xanthotoxin
299-42-3	(-)-ephedrine
305-01-1	esculetin
327-97-9	chlorogenic acid
331-39-5	caffeic acid
437-64-9	genkwanin
446-72-0	genistein
454-14-8	cuscohygrine
458-37-7	curcumin
463-40-1	linolenic acid
464-92-6	asiatic acid
464-98-2	γ-taraxasterol
468-28-0	lupulone
469-32-9	hamamelitannin
470-82-6	1,8-cineole
471-53-4	glycyrrhetic acid
471-66-9	α-boswellic acid
472-15-1	betulinic acid
474-58-8	daucosterol
476-66-4	ellagic acid
477-32-7	visnadin
478-42-2	khellinol
478-43-3	rhein
478-61-5	berbamine
478-79-5	khellol
479-91-4	casticin
479-98-1	aucubin
480-10-4	astragalin
480-18-2	taxifolin
480-19-3	isorhamnetin
481-46-9	ginkgetin
481-72-1	aloe-emodin
481-74-3	chrysophanol
482-36-0	hyperoside
482-44-0	imperatorin
483-17-0	cephaeline
483-18-1	emetine
484-20-8	bergapten

CAS RN	Name
485-72-3	formononetin
486-21-5	isofraxidin
486-66-8	daidzein
486-84-0	harman
489-49-6	cetraric acid
489-50-9	fumarprotocetraric acid
490-46-0	*epi*-catechin
491-60-1	frangula emodin anthrone
491-67-8	baicalein
491-80-5	biochanin A
492-39-7	(+)-norpseudoephedrine
492-41-1	(-)-norephedrine
495-60-3	zingiberene
495-85-2	methysticin
497-76-7	arbutin
498-02-2	apocynine
499-75-2	carvacrol
500-62-9	yangonin
500-64-1	kawain
506-26-3	γ-linolenic acid
506-51-4	tetracosanol
507-70-0	borneol
508-02-1	oleanolic acid
509-80-8	mitraphylline
518-82-1	frangula emodin
519-54-0	neocarthamin
520-36-5	apigenin
521-32-4	bilobetin
521-34-6	sciadopitysin
521-61-9	physcione
521-62-0	frangulin A
522-12-3	quercitrin
523-01-3	*O*-methylpsychotrine
523-47-7	β-cadinene
528-43-8	magnolol
528-58-5	cyanidin
529-05-5	chamazulene
529-44-2	myricetin
532-65-0	*ar*-turmerone
535-83-1	trigonelline
537-73-5	(*E*)-isoferulic acid
539-86-6	allicin
545-46-0	uvaol

CAS RN	Name
548-04-9	hypericin
548-19-6	isoginkgetin
550-37-8	valerophenone-*o*-carboxylic acid
551-08-6	(*Z*)-butylidenephtalide
551-15-5	liquiritin
552-70-5	pseudopelletierine
552-79-4	(-)-methylephedrine
556-27-4	alliin
559-74-0	friedelin
562-74-3	terpinen-4-ol
578-86-9	liquiritigenin
587-63-3	dihydrokawain
607-80-7	(+)-sesamin
630-94-4	corynoxeine
631-69-6	β-boswellic acid
632-85-9	wogonin
638-95-9	α-amyrin
661-19-8	docosanol
668-10-0	ammiol
961-29-5	isoliquiritigenin
1058-61-3	sitosterone
1059-14-9	taraxasterol
1135-24-6	(*E*)-ferulic acid
1195-79-5	fenchone
1259-86-5	glaucarubinone
1405-86-3	glycyrrhizin
1415-73-2	aloin A
1448-96-0	(+)-protolichesterinic
1617-53-4	amentoflavone
1898-13-1	cembrene
2050-87-5	diallyl trisulfide
2086-83-1	berberine
2086-96-6	β-canadine
2141-09-5	magnoflorine
2179-57-9	diallyl disulfide
2216-51-5	menthol
2222-07-3	cucurbitacin I
2380-78-1	homovanillyl alcohol
2415-24-9	catalpol
2858-66-4	pelletierine
3338-55-4	*cis*-β-ocimene
3387-41-5	sabinene
3486-67-7	palmatine

CAS RN	Name
3569-10-6	valerenic acid
3621-36-1	columbamine
3621-38-3	jatrorrhizine
3681-93-4	vitexin
3779-59-7	cimigenol
3877-86-9	cucurbitacin D
4128-17-0	farnesyl acetate
4176-16-3	valerenal
4180-23-8	*trans*-anethole
4221-98-1	(-)-α-phellandrene
4261-42-1	iso-orientin
4373-41-5	maslinic acid
4396-01-4	isopelletierine
4431-01-0	(*Z*)-ligustilide
4697-68-1	speciophylline
4963-01-3	isomitraphylline
5041-81-6	isoliquiritin
5092-09-1	saikogenin A
5096-57-1	α-canadine
5119-48-2	withaferin A
5171-37-9	isopteropodine
5373-11-5	cynaroside
5508-58-7	andrographolide
5573-16-0	saikogenin D
5629-60-7	pteropodine
5892-54-6	γ-crocetin
5939-57-1	cucurbitacin F
5945-86-8	nimbin
5948-04-9	dihydrocarvone
5957-80-2	carnosol
5968-70-7	3-*O*-acetyl-β-boswellic acid
5989-27-5	(+)-limonene
6199-67-3	cucurbitacin B
6537-80-0	chicoric acid
6753-98-6	humulene (α-caryophyllene)
6754-13-8	helenalin
6754-58-1	xanthohumol
6801-40-7	magnocurarine
6807-92-7	cucurbitine
6853-87-8	methylcysteine *S*-oxide
6859-01-4	isorhynchophylline
6873-13-8	phellodendrine
6906-38-3	delphinidin 3-monoglucoside

CAS RN	Name
6906-39-4	peonidin 3-monoglucoside
6926-08-5	harpagide
6988-81-4	petunidin 3-monoglucoside
7084-24-4	cyanidin 3-monoglucoside
7228-78-6	malvidin 3-monoglucoside
7374-79-0	eleutheroside E1
7432-28-2	schisandrol A
7633-29-6	psychotrine
7785-54-8	(+)-α-terpinyl acetate
8063-16-9	psyllium
10219-75-7	eremophilene
11006-91-0	aloinoside B
11021-13-9	ginsenoside Rb2
11021-14-0	ginsenoside Rc
11027-63-7	agnuside
11079-53-1	hyperforin
11137-63-6	sennoside E
11141-17-6	azadirachtin
13844-01-4	barringtogenol C
14019-66-0	uncarine F
14062-59-0	glucofrangulin B
14073-97-3	menthone
14101-04-3	frangulin B
14215-86-2	sweroside
14356-59-3	saikogenin F
14907-98-3	brusatol
14965-20-9	chrysosplenol D
15291-75-5	ginkgolide A
15291-76-6	ginkgolide C
15291-77-7	ginkgolide B
15291-78-8	ginkgolide M
15345-89-8	demethoxyyangonin
15401-60-2	ipecoside
15486-24-5	eleutheroside C
15514-06-4	procyanidin
16830-15-2	asiaticoside
16845-16-2	eleutheroside B1
17019-92-0	11-oxo-β-boswellic acid
17226-75-4	khellinin
17795-24-3	propylcysteine *S*-oxide
17910-09-7	curzerene
18175-79-6	saikogenin G
18234-46-3	baldrinal
18262-45-8	(-)-11-deoxyaloin
18296-44-1	valtrate
18444-66-1	cucurbitacin E
18449-41-7	madecassic acid
18609-16-0	deacetylnimbin
18642-44-9	actein
18747-42-7	*N*-methylisopelletierine
18942-26-2	chebulinic acid
19210-12-9	harpagoside
19467-03-9	melittoside
19533-92-7	alphitolic acid
19666-76-3	panaxadiol
19764-02-4	fragilin
19902-91-1	dihydromethysticin
19912-61-9	(5*E*,9*Z*)-furanodiene
20229-56-5	spiraeoside
20283-92-5	rosmarinic acid
20315-25-7	procyanidin B1
20347-65-3	bornyl acetate
20486-27-5	procumbide
20493-56-5	curzerenone
20554-84-1	parthenolide
20633-72-1	monomelittoside
20633-84-5	scolymoside
20675-96-1	syringenin
20736-09-8	saikosaponin A
20831-76-9	gentiopicroside
20853-07-0	protoaescigenin
20874-52-6	saikosaponin D
21018-84-8	amarogentin
21133-53-9	glucofrangulin A
21453-69-0	(+)-syringaresinol
21499-66-1	brucein D
21637-25-2	isoquercitrin
21967-41-9	baicalin
22255-13-6	guajaverin
22427-39-0	ginsenoside Rg1
22608-11-3	desmethoxycurcumin
22888-70-6	silybin
23089-26-1	(-)-α-bisabolol
23094-71-5	chebulagic acid
23180-57-6	paeoniflorin
23513-08-8	[8]-gingerol

CAS RN	Name
23513-13-5	[6]-shogaol
23513-14-6	[6]-gingerol
23513-15-7	[10]-gingerol
23567-23-9	procyanidin B3
23666-13-9	vicenin-2
24815-24-5	rescinnamine
24959-84-0	canin
25161-41-5	acevaltrate
25368-09-6	β-crocetin
25514-29-8	brucein B
25514-30-1	brucein C
25514-31-2	brucein A
26020-14-4	calenduloside E
26020-29-1	calenduloside H
26472-41-3	humulone
27208-74-8	acteol
27215-14-1	neoandrographolide
27409-30-9	picroside I
27876-94-4	α-crocetin
28371-16-6	aloin B
28608-75-5	orientin
29106-49-8	procyanidin B2
29106-51-2	procyanidin B4
29428-58-8	lucenin-2
29431-84-3	artecanin
29782-68-1	silydianin
29836-41-7	salicortin
29883-15-6	amygdalin
30655-48-2	withanolide D
30964-13-7	cynarin
31078-10-1	isovaltrate
32725-74-9	calenduloside A
32791-84-7	panaxatriol
33171-05-0	bisdesmethoxycurcumin
33570-04-6	bilobalide
33889-69-9	silychristin
34257-95-9	11α,13-dihydrohelenalin
34366-31-9	senegin II
34540-22-2	madecassoside
35354-74-6	honokiol
35730-78-0	cynaropicrin
35906-36-6	(*E*)-senegin III
35988-27-3	kutkoside
36338-96-2	carthamin
37271-16-2	sennoside C
37271-17-3	sennoside D
37719-98-5	angelicone
38953-85-4	isovitexin
39012-20-9	picroside II
39025-23-5	(*Z*)-guggulsterone
39025-24-6	(*E*)-guggulsterone
39025-25-7	guggulsterol I
39025-26-8	guggulsterol-II
39025-27-9	guggulsterol-III
39432-56-9	eleutheroside E
41451-75-6	bruceantin
41743-67-3	[3]-gingerol
41743-68-4	[4]-gingerol
41743-69-5	[5]-gingerol
41753-43-9	ginsenoside Rb1
41943-03-7	mukulol
42553-65-1	A-crocin
51014-29-0	isocorynoxeine
51018-28-1	(+)-methylpseudoephedrine
51415-02-2	calenduloside F
51938-32-0	schaftoside
52012-29-0	isoschaftoside
52286-58-5	ginsenoside Rf
52286-59-6	ginsenoside Re
52286-74-5	ginsenoside Rg2
52705-93-8	ginsenoside Rd
52842-23-6	sennoside F
52949-83-4	ajugol
53472-37-0	3′-methylmyricetin
53683-73-1	ailanthinone
53823-08-8	cascaroside A
53823-09-9	cascaroside C
53861-34-0	cascaroside B
53861-35-1	cascaroside D
53962-20-2	patrinoside
55056-80-9	protodioscin
55466-05-2	jujuboside B
55750-84-0	B-crocin
55750-85-1	C-crocin
55903-92-9	cucurbitacin R
55954-61-5	pseudohypericin

CAS RN	Name
56645-88-6	aloinoside A
57710-64-2	D-crocin
58050-17-2	E-crocin
58316-41-9	saikosaponin B2
58316-42-0	saikosaponin B3
58479-68-8	platycodin D
58546-54-6	schisandrol B
58558-08-0	saikosaponin B1
58558-09-1	saikosaponin B4
59440-97-0	(+)-echinolone
61263-49-8	vitexilactone
61281-37-6	schisandrin B
61281-38-7	schisandrin A
62488-53-3	3-vinyl-1,2-dithiin
63644-62-2	coniferyl ferulate
64461-95-6	picroside III
64820-99-1	vitexin 2´´-rhamnoside
65995-63-3	punicalagin
65995-64-4	punicalin
66176-93-0	cimicifugoside
66663-90-9	platycodin D2
66663-91-0	polygalacin D
66663-92-1	polygalacin D2
66779-34-8	platycodin A
66779-35-9	platycodin C
67416-61-9	3-*O*-acetyl-11-oxo-β-boswellic acid
67910-07-0	homobaldrinal
69176-52-9	gomisin N
69884-00-0	pseudoginsenoside F11
69980-49-0	(+)-11-deoxyaloin
71013-41-7	valtroxal
71616-00-7	achillicin
73543-67-6	α-dimorphecolic acid
73889-54-0	chrysaloin
75911-12-5	taraxacolide glucoside
75911-14-7	taraxinic acid glucoside
75917-90-7	(10*E*)-*N*-isobutyldodecatetraenamide
75991-58-1	(3*S*,9*R*)-tetrahydroridentin B
77053-35-1	5´-methoxybilobetin
77448-63-6	(10*Z*)-*N*-isobutyldodecatetraenamide
77790-55-7	neoolivil
77943-54-5	zizyphus saponin III
77943-56-7	zizyphus saponin I
77943-83-0	zizyphus saponin II
78281-02-4	hydroxysafflor yellow A
79484-75-6	eleutheroside D
79974-46-2	tribulosin
80028-57-5	vinyl-1,3-dithiin (2-)
80151-77-5	(-)-tussilagine
81348-81-4	momordicoside F1
81348-82-5	momordicoside F2
81348-83-6	momordicoside L
81348-84-7	momordicoside K
81371-54-2	momordicoside G
81371-55-3	momordicoside I
81720-05-0	rehmannioside A
81720-06-1	rehmannioside B
81720-07-2	rehmannioside C
81720-08-3	rehmannioside D
82209-72-1	andropanoside
82209-76-5	andrographiside
82508-14-3	β-turmerone
82508-15-4	α-turmerone
82854-37-3	echinacoside
82901-10-8	(5*E*,8*S*,9*E*)-2-methoxyfuranodiene
83199-38-6	angelol G
84638-55-1	acetoxyvalerenic acid
84676-88-0	isoastragaloside I
84676-89-1	astragaloside II
84680-75-1	astragaloside I
84687-42-3	astragaloside III
84687-43-4	astragaloside IV
84687-44-5	astragaloside V
84687-45-6	astragaloside VI
84687-46-7	astragaloside VII
85532-77-0	safflor yellow A
86764-11-6	isoastragaloside II
87605-93-4	furanoeudesma-1,3-diene
88010-65-5	(9*E*)-4,5-dihydrofuranodien-6-one
89913-60-0	3-*O*-acetyl-α-boswellic acid

CAS RN	Name
91108-32-6	isotussilagine
91574-92-4	safflor yellow B
92284-99-6	(*E*)-ajoene
92285-00-2	(*Z*)-ajoene
92935-94-9	angelicide
98449-40-2	taraxacoside
99742-03-7	[1]-gingerol
99742-07-1	[7]-shogaol
99742-08-2	[9]-shogaol
99742-09-3	[11]-shogaol
99742-10-6	[12]-shogaol
101927-24-6	docosyl (*E*)-ferulate
101927-25-7	tetracosyl (*E*)-ferulate
104404-17-3	ceramides
107438-79-9	ginkgolide J
108376-98-3	(5*E*,8*R*,9*E*)-2-methoxyfuranodiene
111545-28-9	rheinoside A
111545-29-0	rheinoside C
111614-10-9	rheinoside B
111614-11-0	rheinoside D
111957-48-3	kaempferol diglycoside coumarate
126038-52-6	zwiebelanes
128748-74-8	uncarinic acid B
143061-65-8	quercetin diglycoside coumarate
143183-63-5	adhyperforin
149475-43-4	tinctormine
162613-71-0	(*Z*)-senegasaponin B
162613-72-1	(*Z*)-senegasaponin A
162681-52-9	(*Z*)-senegin II
162681-53-0	(*Z*)-senegin III
162762-97-2	(*E*)-senegasaponin A
162870-58-8	(*E*)-senegasaponin B
163589-51-3	desacylsenegin II
164178-32-9	cascaroside E
164322-83-2	cascaroside F
166833-80-3	chebulanin
173557-02-3	desacylsenegasaponin A
180572-65-0	desacylsenegin III
194413-09-7	withasomniferol A
206256-62-4	uncarinic acid A
218622-84-5	tribulusamide A
218622-86-7	tribulusamide B
311310-49-3	tribulosaponin A
311310-52-8	tribulosaponin B
333332-41-5	goyaglycoside a
333332-48-2	goyaglycoside b
333332-49-3	goyaglycoside c
333332-50-6	goyaglycoside d
333333-12-3	goyaglycoside e
333333-13-4	goyaglycoside f
333333-14-5	goyaglycoside g
333333-19-0	goyaglycoside h
333333-20-3	goyasaponin I
333333-23-6	goyasaponin II
333333-27-0	goyasaponin III
360762-35-2	*N*-isobutyldodecatetraenamide
459167-05-6	vitexlactam A

Annex 7

Cumulative index of major chemical constituents (*ordered by molecular formula*)

Cumulative index of major chemical compounds whose chemical structure drawings were presented in four volumes of WHO monographs on selected medicinal plants

Mol. form.	Name
C-4 H-9 N O-3 S	methylcysteine *S*-oxide
C-5 H-10 N-2 O-2	cucurbitine
C-5 H-10 O	2-methylbut-3-en-2-ol
C-6 H-6 O-2	hydroquinone
C-6 H-6 O-3	maltol
C-6 H-8 S-2	2-vinyl-1,3-dithiin
C-6 H-8 S-2	3-vinyl-1,2-dithiin
C-6 H-10 O S-2	allicin
C-6 H-10 O S-2	zwiebelanes
C-6 H-10 S-2	diallyl disulfide
C-6 H-10 S-3	diallyl trisulfide
C-6 H-11 N O-3 S	alliin
C-6 H-11 N O-3 S	1-propenylcysteine oxide
C-6 H-13 N O-3 S	propylcysteine *S*-oxide
C-7 H-6 O-2	*p*-hydroxybenzaldehyde
C-7 H-6 O-5	gallic acid
C-7 H-7 N O-2	trigonelline
C-8 H-8 O-2	*p*-anisaldehyde
C-8 H-8 O-4	vanillic acid
C-8 H-15 N O	isopelletierine
C-8 H-15 N O	pelletierine
C-8 H-16 O-6	eleutheroside C
C-9 H-6 O-2	coumarin
C-9 H-6 O-3	umbelliferone
C-9 H-6 O-4	esculetin
C-9 H-8 O	cinnamaldehyde
C-9 H-8 O-4	caffeic acid
C-9 H-10 O-3	apocynine
C-9 H-12 O-3	homovanillyl alcohol
C-9 H-13 N O	(-)-norephedrine
C-9 H-13 N O	(+)-norpseudoephedrine
C-9 H-14 O S-3	(*E*)-ajoene
C-9 H-14 O S-3	(*Z*)-ajoene
C-9 H-15 N O	pseudopelletierine
C-9 H-17 N O	*N*-methylisopelletierine
C-10 H-8 O-4	scopoletin
C-10 H-10 O-4	(*E*)-ferulic acid
C-10 H-10 O-4	(*E*)-isoferulic acid
C-10 H-12 O	*trans*-anethole
C-10 H-12 O	estragole
C-10 H-12 O	methylchavicol
C-10 H-12 O-2	eugenol
C-10 H-14	*p*-cymene
C-10 H-14 O	carvacrol
C-10 H-14 O	carvone
C-10 H-14 O	safranal
C-10 H-14 O	thymol
C-10 H-15 N O	(-)-ephedrine
C-10 H-15 N O	(+)-pseudoephedrine

Mol. form.	Name
C-10 H-16	(+)-limonene
C-10 H-16	*cis*-β-ocimene
C-10 H-16	(-)-α-phellandrene
C-10 H-16	α-pinene
C-10 H-16	β-pinene
C-10 H-16	sabinene
C-10 H-16	α-terpinene
C-10 H-16	γ-terpinene
C-10 H-16 O	camphor
C-10 H-16 O	dihydrocarvone
C-10 H-16 O	fenchone
C-10 H-16 O	geranial
C-10 H-16 O	neral
C-10 H-17 N O-3	isotussilagine
C-10 H-17 N O-3	(-)-tussilagine
C-10 H-18 O	borneol
C-10 H-18 O	1,8-cineole
C-10 H-18 O	citronellal
C-10 H-18 O	geraniol
C-10 H-18 O	linalool (β-linalool)
C-10 H-18 O	menthone
C-10 H-18 O	nerol
C-10 H-18 O	terpinen-4-ol
C-10 H-18 O	α-terpineol
C-10 H-20 O	menthol
C-11 H-10 O-5	isofraxidin
C-11 H-14 O-2	methyleugenol
C-11 H-14 O-4	syringenin
C-11 H-17 N O	(-)-methylephedrine
C-11 H-17 N O	(+)-methylpse-udoephedrine
C-12 H-8 O-4	bergapten
C-12 H-8 O-4	xanthotoxin
C-12 H-10 N-2	harman
C-12 H-10 O-4	baldrinal
C-12 H-12 O-2	(*Z*)-butylidenephtalide
C-12 H-14 O-2	(*Z*)-ligustilide
C-12 H-14 O-3	eugenol acetate
C-12 H-14 O-3	valerophenone-*o*-carboxylic acid
C-12 H-16 O-4	[1]-gingerol
C-12 H-16 O-7	arbutin
C-12 H-20 O-2	bornyl acetate
C-12 H-20 O-2	linalyl acetate
C-12 H-20 O-2	(+)-α-terpinyl acetate
C-12 H-24 O-2	lauric acid
C-13 H-10 O-4	visnagin
C-13 H-10 O-5	khellinol
C-13 H-10 O-5	khellol
C-13 H-18 O-7	salicin
C-13 H-24 N-2 O	cuscohygrine
C-14 H-6 O-8	ellagic acid
C-14 H-12 O-3	demethoxyyangonin
C-14 H-12 O-5	khellin
C-14 H-12 O-6	ammiol
C-14 H-14 O-3	kawain
C-14 H-16	chamazulene
C-14 H-16 O-3	dihydrokawain
C-14 H-16 O-8	bilobalide
C-14 H-20 O-4	[3]-gingerol
C-14 H-24 O-2	(+)-echinolone
C-15 H-8 O-6	rhein
C-15 H-10 O-4	chrysophanol
C-15 H-10 O-4	daidzein
C-15 H-10 O-5	aloe-emodin
C-15 H-10 O-5	apigenin
C-15 H-10 O-5	baicalein
C-15 H-10 O-5	frangula emodin
C-15 H-10 O-5	genistein
C-15 H-10 O-7	quercetin
C-15 H-10 O-8	myricetin
C-15 H-11 Cl O-6	cyanidin
C-15 H-12 O-4	frangula emodin anthrone
C-15 H-12 O-4	isoliquiritigenin
C-15 H-12 O-4	liquiritigenin
C-15 H-12 O-7	taxifolin
C-15 H-14 O-4	yangonin
C-15 H-14 O-5	methysticin
C-15 H-14 O-6	catechin
C-15 H-14 O-6	*epi*-catechin
C-15 H-16 O-4	homobaldrinal
C-15 H-16 O-5	dihydromethysticin
C-15 H-18 O	furanoeudesma-1,3-diene
C-15 H-18 O-2	curzerenone

Mol. form.	Name
C-15 H-18 O-4	helenalin
C-15 H-18 O-5	artecanin
C-15 H-18 O-5	canin
C-15 H-20 O	curzerene
C-15 H-20 O	(5*E*,9*Z*)-furanodiene
C-15 H-20 O	*ar*-turmerone
C-15 H-20 O-2	(9*E*)-4,5-dihydro-furanodien-6-one
C-15 H-20 O-3	parthenolide
C-15 H-20 O-4	11α,13-dihydro-helenalin
C-15 H-20 O-4	3β-hydroxy-parthenolide
C-15 H-20 O-8	fragilin
C-15 H-22 O	α-turmerone
C-15 H-22 O	β-turmerone
C-15 H-22 O	valerenal
C-15 H-22 O-2	valerenic acid
C-15 H-22 O-4	[4]-gingerol
C-15 H-22 O-9	aucubin
C-15 H-22 O-10	catalpol
C-15 H-22 O-10	monomelittoside
C-15 H-22 O-10	procumbide
C-15 H-24	β-cadinene
C-15 H-24	β-caryophyllene
C-15 H-24	eremophilene
C-15 H-24	humulene (α-caryophyllene)
C-15 H-24	zingiberene
C-15 H-24 O-4	(3*S*,9*R*)-tetrahydroridentin B
C-15 H-24 O-9	ajugol
C-15 H-24 O-10	harpagide
C-15 H-26 O	(-)-α-bisabolol
C-15 H-26 O-2	bornyl isovalerate
C-16 H-12 O-4	formononetin
C-16 H-12 O-5	biochanin A
C-16 H-12 O-5	genkwanin
C-16 H-12 O-5	physcione
C-16 H-12 O-5	wogonin
C-16 H-12 O-7	isorhamnetin
C-16 H-12 O-8	3´-methylmyricetin
C-16 H-13 Cl O-6	peonidin
C-16 H-14 O-4	imperatorin
C-16 H-16 O-5	angelicone
C-16 H-18 O-9	chlorogenic acid
C-16 H-20 O-9	gentiopicroside
C-16 H-22 O-2	(5*E*,8*R*,9*E*)-2-methoxy-furanodiene
C-16 H-22 O-2	(5*E*,8*S*,9*E*)-2-methoxy-furanodiene
C-16 H-22 O-9	sweroside
C-16 H-24 O-4	[5]-gingerol
C-16 H-25 N O	*N*-isobutyldodecatetraenamide
C-16 H-25 N O	(10*E*)-*N*-isobutyldodecatetraenamide
C-16 H-25 N O	(10*Z*)-*N*-isobutyldodecatetraenamide
C-16 H-26 O-7	picrocrocin
C-17 H-20 O-10	eleutheroside B1
C-17 H-22 O-5	achillicin
C-17 H-22 O-7	valtroxal
C-17 H-24 O-3	[6]-shogaol
C-17 H-24 O-4	acetoxyvalerenic acid
C-17 H-24 O-9	eleutheroside B
C-17 H-26 O-4	[6]-gingerol
C-17 H-28 O-2	farnesyl acetate
C-18 H-16 O-8	chrysosplenol D
C-18 H-16 O-8	rosmarinic acid
C-18 H-18 O-2	honokiol
C-18 H-18 O-2	magnolol
C-18 H-22 O-10	taraxacoside
C-18 H-25 N O-5	senecionine
C-18 H-26 O-3	[7]-shogaol
C-18 H-30 O-2	linolenic acid
C-18 H-30 O-2	γ-linolenic acid
C-18 H-32 O-2	linoleic acid
C-18 H-32 O-3	α-dimorphecolic acid
C-18 H-34 O-2	oleic acid
C-18 H-34 O-3	ricinoleic acid
C-19 H-16 O-4	bisdesmethoxycurcumin
C-19 H-18 O-8	casticin
C-19 H-20 O-10	khellinin
C-19 H-22 O-6	cynaropicrin
C-19 H-24 N O-3	magnocurarine

Mol. form.	Name
C-19 H-30 O-4	[8]-gingerol
C-19 H-32 O-4	(+)-protolichesterinic acid
C-20 H-18 N O-4	berberine
C-20 H-18 O-5	desmethoxycurcumin
C-20 H-18 O-6	(+)-sesamin
C-20 H-18 O-9	cetraric acid
C-20 H-18 O-9	frangulin B
C-20 H-18 O-11	guajaverin
C-20 H-20 N O-4	columbamine
C-20 H-20 N O-4	jatrorrhizine
C-20 H-20 O-6	coniferyl ferulate
C-20 H-20 O-14	hamamelitannin
C-20 H-21 N O-4	α-canadine
C-20 H-21 N O-4	β-canadine
C-20 H-24 N O-4	magnoflorine
C-20 H-24 N O-4	phellodendrine
C-20 H-24 O-4	α-crocetin
C-20 H-24 O-7	angelol G
C-20 H-24 O-7	neoolivil
C-20 H-24 O-9	ginkgolide A
C-20 H-24 O-10	ginkgolide B
C-20 H-24 O-10	ginkgolide J
C-20 H-24 O-10	ginkgolide M
C-20 H-24 O-10	salicortin
C-20 H-24 O-11	ginkgolide C
C-20 H-26 O-4	carnosol
C-20 H-26 O-9	brucein D
C-20 H-30 O-3	[9]-shogaol
C-20 H-30 O-5	andrographolide
C-20 H-32	cembrene
C-20 H-34 O	mukulol
C-21 H-18 O-11	baicalin
C-21 H-20 O-6	curcumin
C-21 H-20 O-9	frangulin A
C-21 H-20 O-10	isovitexin
C-21 H-20 O-10	vitexin
C-21 H-20 O-11	astragalin
C-21 H-20 O-11	cynaroside
C-21 H-20 O-11	iso-orientin
C-21 H-20 O-11	orientin
C-21 H-20 O-11	quercitrin
C-21 H-20 O-12	hyperoside
C-21 H-20 O-12	isoquercitrin
C-21 H-20 O-12	spiraeoside
C-21 H-21 Cl O-11	cyanidin 3-mono-glucoside
C-21 H-21 Cl O-12	delphinidin 3-monoglucoside
C-21 H-21 N O-6	hydrastine
C-21 H-22 N O-4	palmatine
C-21 H-22 O-5	xanthohumol
C-21 H-22 O-8	chrysaloin
C-21 H-22 O-8	(-)-11-deoxyaloin
C-21 H-22 O-8	(+)-11-deoxyaloin
C-21 H-22 O-9	aloin A
C-21 H-22 O-9	aloin B
C-21 H-22 O-9	isoliquiritin
C-21 H-22 O-9	liquiritin
C-21 H-22 O-11	neocarthamin
C-21 H-24 N-2 O-4	isomitraphylline
C-21 H-24 N-2 O-4	isopteropodine
C-21 H-24 N-2 O-4	mitraphylline
C-21 H-24 N-2 O-4	pteropodine
C-21 H-24 N-2 O-4	speciophylline
C-21 H-24 N-2 O-4	uncarine F
C-21 H-24 O-7	visnadin
C-21 H-26 O-4	β-crocetin
C-21 H-27 N O-11	amygdalin
C-21 H-28 O-2	(*E*)-guggulsterone
C-21 H-28 O-2	(*Z*)-guggulsterone
C-21 H-28 O-9	taraxinic acid glucoside
C-21 H-30 O-5	humulone
C-21 H-32 O-9	taraxacolide glucoside
C-21 H-32 O-15	melittoside
C-21 H-32 O-15	rehmannioside A
C-21 H-32 O-15	rehmannioside B
C-21 H-34 O-4	[10]-gingerol
C-21 H-34 O-11	patrinoside
C-21 H-34 O-14	rehmannioside C
C-22 H-16 O-12	fumarprotocetraric acid
C-22 H-18 O-12	chicoric acid
C-22 H-23 Cl O-11	peonidin 3-mono-glucoside
C-22 H-23 Cl O-12	petunidin 3-mono-glucoside

Mol. form.	Name
C-22 H-26 N-2 O-4	corynoxeine
C-22 H-26 N-2 O-4	isocorynoxeine
C-22 H-26 O-8	(+)-syringaresinol
C-22 H-26 O-11	2´-*O*-acetylsalicortin
C-22 H-26 O-11	agnuside
C-22 H-28 N-2 O-4	isorhynchophylline
C-22 H-28 N-2 O-4	rhynchophylline
C-22 H-28 O-4	γ-crocetin
C-22 H-28 O-6	quassin
C-22 H-30 O-8	isovaltrate
C-22 H-30 O-8	valtrate
C-22 H-34 O-3	[11]-shogaol
C-22 H-34 O-5	vitexilactone
C-22 H-35 N O-4	vitexlactam A
C-22 H-46 O	docosanol
C-23 H-25 Cl O-12	malvidin 3-mono-glucoside
C-23 H-28 O-6	gomisin N
C-23 H-28 O-6	schisandrin B
C-23 H-28 O-7	schisandrol B
C-23 H-28 O-11	brucein B
C-23 H-28 O-11	paeoniflorin
C-23 H-28 O-13	kutkoside
C-23 H-28 O-13	picroside II
C-23 H-36 O-3	[12]-shogaol
C-24 H-28 O-4	angelicide
C-24 H-28 O-11	picroside I
C-24 H-30 O-11	harpagoside
C-24 H-32 O-6	schisandrin A
C-24 H-32 O-7	schisandrol A
C-24 H-32 O-10	acevaltrate
C-24 H-50 O	tetracosanol
C-25 H-22 O-10	silybin
C-25 H-22 O-10	silychristin
C-25 H-22 O-10	silydianin
C-25 H-24 O-12	cynarin
C-25 H-30 O-13	picroside III
C-25 H-34 O-9	ailanthinone
C-25 H-34 O-10	glaucarubinone
C-26 H-28 O-14	glucofrangulin B
C-26 H-28 O-14	isoschaftoside
C-26 H-28 O-14	schaftoside
C-26 H-32 O-12	brusatol
C-26 H-34 O-9	E-crocin
C-26 H-34 O-11	brucein A
C-26 H-38 O-4	lupulone
C-26 H-40 O-8	neoandrographolide
C-26 H-40 O-9	andropanoside
C-26 H-40 O-10	andrographiside
C-27 H-24 O-19	chebulanin
C-27 H-30 O-14	glucofrangulin A
C-27 H-30 O-14	vitexin 2´´-rhamnoside
C-27 H-30 O-15	safflor yellow A
C-27 H-30 O-15	scolymoside
C-27 H-30 O-15	vicenin-2
C-27 H-30 O-16	lucenin-2
C-27 H-30 O-16	rheinoside C
C-27 H-30 O-16	rheinoside D
C-27 H-30 O-16	rutin
C-27 H-30 O-17	rheinoside A
C-27 H-30 O-17	rheinoside B
C-27 H-31 N O-14	tinctormine
C-27 H-32 O-13	aloinoside A
C-27 H-32 O-13	aloinoside B
C-27 H-32 O-13	cascaroside C
C-27 H-32 O-13	cascaroside D
C-27 H-32 O-14	cascaroside A
C-27 H-32 O-14	cascaroside B
C-27 H-32 O-14	cascaroside E
C-27 H-32 O-14	cascaroside F
C-27 H-32 O-16	hydroxysafflor yellow A
C-27 H-35 N O-12	ipecoside
C-27 H-42 O-20	rehmannioside D
C-27 H-44 O-3	guggulsterol-III
C-27 H-44 O-4	guggulsterol I
C-27 H-46 O-3	guggulsterol-II
C-28 H-34 O-8	deacetylnimbin
C-28 H-36 N-2 O-4	psychotrine
C-28 H-36 O-11	bruceantin
C-28 H-36 O-13	brucein C
C-28 H-36 O-13	eleutheroside E1
C-28 H-38 N-2 O-4	cephaeline
C-28 H-38 O-6	withaferin A
C-28 H-38 O-6	withanolide D
C-28 H-38 O-7	withasomniferol A

Mol. form.	Name
C-29 H-30 O-13	amarogentin
C-29 H-38 N-2 O-4	*O*-methylpsychotrine
C-29 H-40 N-2 O-4	emetine
C-29 H-48 O	sitosterone
C-29 H-50 O	β-sitosterol
C-30 H-16 O-8	hypericin
C-30 H-16 O-9	pseudohypericin
C-30 H-18 O-10	amentoflavone
C-30 H-26 O-12	procyanidin
C-30 H-26 O-12	procyanidin B1
C-30 H-26 O-12	procyanidin B2
C-30 H-26 O-12	procyanidin B3
C-30 H-26 O-12	procyanidin B4
C-30 H-26 O-14	proanthocyanidins
C-30 H-26 O-14	prodelphinidin
C-30 H-36 O-9	nimbin
C-30 H-42 O-7	cucurbitacin I
C-30 H-44 O-7	cucurbitacin D
C-30 H-46 O-4	glycyrrhetic acid
C-30 H-46 O-4	11-oxo-β-boswellic acid
C-30 H-46 O-6	acteol
C-30 H-46 O-7	cucurbitacin F
C-30 H-46 O-7	cucurbitacin R
C-30 H-48 O-3	betulinic acid
C-30 H-48 O-3	α-boswellic acid
C-30 H-48 O-3	β-boswellic acid
C-30 H-48 O-3	oleanolic acid
C-30 H-48 O-3	ursolic acid
C-30 H-48 O-4	alphitolic acid
C-30 H-48 O-4	maslinic acid
C-30 H-48 O-4	saikogenin A
C-30 H-48 O-4	saikogenin D
C-30 H-48 O-4	saikogenin F
C-30 H-48 O-4	saikogenin G
C-30 H-48 O-5	asiatic acid
C-30 H-48 O-5	cimigenol
C-30 H-48 O-6	madecassic acid
C-30 H-50	squalene
C-30 H-50 O	α-amyrin
C-30 H-50 O	friedelin
C-30 H-50 O	taraxasterol
C-30 H-50 O	γ-taraxasterol
C-30 H-50 O-2	uvaol
C-30 H-50 O-5	barringtogenol C
C-30 H-50 O-6	protoaescigenin
C-30 H-52 O-3	panaxadiol
C-30 H-52 O-4	panaxatriol
C-31 H-20 O-10	bilobetin
C-32 H-22 O-10	ginkgetin
C-32 H-22 O-10	isoginkgetin
C-32 H-22 O-11	5´-methoxybilobetin
C-32 H-44 O-8	cucurbitacin E
C-32 H-44 O-14	C-crocin
C-32 H-44 O-14	D-crocin
C-32 H-46 O-8	cucurbitacin B
C-32 H-48 O-5	3-*O*-acetyl-11-oxo-β-boswellic acid
C-32 H-50 O-4	3-*O*-acetyl-α-boswellic acid
C-32 H-50 O-4	3-*O*-acetyl-β-boswellic acid
C-32 H-54 O-4	docosyl (*E*)-ferulate
C-33 H-24 O-10	sciadopitysin
C-33 H-40 N-2 O-9	reserpine
C-34 H-22 O-22	punicalin
C-34 H-46 O-18	eleutheroside D
C-34 H-46 O-18	eleutheroside E
C-34 H-58 O-4	tetracosyl (*E*)-ferulate
C-35 H-42 N-2 O-9	rescinnamine
C-35 H-44 O-16	azadirachtin
C-35 H-46 O-20	echinacoside
C-35 H-52 O-4	hyperforin
C-35 H-60 O-6	daucosterol
C-36 H-34 N-2 O-9	tribulusamide B
C-36 H-36 N-2 O-8	tribulusamide A
C-36 H-36 O-17	kaempferol diglycoside coumarate
C-36 H-36 O-18	quercetin diglycoside coumarate
C-36 H-54 O-4	adhyperforin
C-36 H-56 O-9	calenduloside E
C-36 H-58 O-8	momordicoside F2
C-36 H-58 O-8	momordicoside I
C-36 H-58 O-9	momordicoside L

Mol. form.	Name
C-37 H-40 N-2 O-6	berbamine
C-37 H-54 O-11	cimicifugoside
C-37 H-56 O-11	actein
C-37 H-60 O-8	momordicoside F1
C-37 H-60 O-8	momordicoside G
C-37 H-60 O-9	goyaglycoside a
C-37 H-60 O-9	goyaglycoside b
C-37 H-60 O-9	momordicoside K
C-38 H-54 O-19	B-crocin
C-38 H-62 O-9	goyaglycoside c
C-38 H-62 O-9	goyaglycoside d
C-40 H-56 O-7	uncarinic acid A
C-40 H-56 O-7	uncarinic acid B
C-41 H-30 O-27	chebulagic acid
C-41 H-32 O-27	chebulinic acid
C-41 H-68 O-14	astragaloside III
C-41 H-68 O-14	astragaloside IV
C-42 H-38 O-20	sennoside A
C-42 H-38 O-20	sennoside B
C-42 H-40 O-19	sennoside C
C-42 H-40 O-19	sennoside D
C-42 H-62 O-16	glycyrrhizin
C-42 H-66 O-14	calenduloside F
C-42 H-68 O-13	calenduloside A
C-42 H-68 O-13	goyaglycoside e
C-42 H-68 O-13	goyaglycoside f
C-42 H-68 O-13	saikosaponin A
C-42 H-68 O-13	saikosaponin B1
C-42 H-68 O-13	saikosaponin B2
C-42 H-68 O-13	saikosaponin D
C-42 H-70 O-15	goyaglycoside h
C-42 H-72 O-13	ginsenoside Rg2
C-42 H-72 O-14	ginsenoside Rf
C-42 H-72 O-14	ginsenoside Rg1
C-42 H-72 O-14	pseudoginsenoside F11
C-43 H-42 O-22	carthamin
C-43 H-70 O-14	goyaglycoside g
C-43 H-70 O-15	astragaloside II
C-43 H-70 O-15	isoastragaloside II
C-43 H-72 O-14	saikosaponin B3
C-43 H-72 O-14	saikosaponin B4
C-44 H-38 O-23	sennoside E
C-44 H-38 O-23	sennoside F
C-44 H-64 O-24	A-crocin
C-45 H-38 O-18	proanthocyanidin trimer
C-45 H-72 O-16	astragaloside I
C-45 H-72 O-16	isoastragaloside I
C-47 H-76 O-17	zizyphus saponin I
C-47 H-76 O-17	zizyphus saponin II
C-47 H-78 O-19	astragaloside V
C-47 H-78 O-19	astragaloside VI
C-47 H-78 O-19	astragaloside VII
C-48 H-28 O-30	punicalagin
C-48 H-54 O-27	safflor yellow B
C-48 H-76 O-19	calenduloside H
C-48 H-78 O-19	asiaticoside
C-48 H-78 O-20	madecassoside
C-48 H-82 O-18	ginsenoside Rd
C-48 H-82 O-18	ginsenoside Re
C-49 H-76 O-19	goyasaponin III
C-51 H-84 O-21	tribulosaponin A
C-51 H-84 O-22	protodioscin
C-51 H-84 O-22	tribulosaponin B
C-52 H-84 O-21	jujuboside B
C-52 H-84 O-21	zizyphus saponin III
C-53 H-90 O-22	ginsenoside Rb2
C-53 H-90 O-22	ginsenoside Rc
C-54 H-92 O-23	ginsenoside Rb1
C-55 H-90 O-25	tribulosin
C-57 H-92 O-27	polygalacin D
C-57 H-92 O-28	platycodin D
C-59 H-94 O-29	desacylsenegin II
C-59 H-94 O-29	platycodin A
C-59 H-94 O-29	platycodin C
C-63 H-102 O-32	polygalacin D2
C-63 H-102 O-33	platycodin D2
C-64 H-102 O-33	desacylsenegasaponin A
C-65 H-102 O-31	goyasaponin I
C-65 H-104 O-33	desacylsenegin III
C-69 H-102 O-31	(*E*)-senegasaponin B
C-69 H-102 O-31	(*Z*)-senegasaponin B

Mol. form.	Name
C-70 H-104 O-32	senegin II
C-70 H-104 O-32	(*Z*)-senegin II
C-70 H-110 O-35	goyasaponin II
C-74 H-110 O-35	(*E*)-senegasaponin A
C-74 H-110 O-35	(*Z*)-senegasaponin A
C-75 H-112 O-35	(*E*)-senegin III
C-75 H-112 O-35	(*Z*)-senegin III

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WHO monographs on selected medicinal plants, Volume 3
(ISBN 978 92 4 154702 4), 2007

WHO monographs on selected medicinal plants, Volume 2
(ISBN 92 4 154537 2), 2002

WHO monographs on selected medicinal plants, Volume 1
(ISBN 92 4 154517 8), 1999

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General Guidelines for Methodologies on Research and Evaluation of Traditional Medicine
(WHO/EDM/TRM/2000.1), 2000

Consumer information:

WHO guidelines on development of consumer information on proper use of traditional medicine and complementary/alternative medicine
(ISBN 92 4 159170 6), 2004

Further information on WHO technical documents in the field of traditional medicine including those listed above, can be found at the address below:
http://www.who.int/medicinedocs/en/cl/CL1.1.1.11/

WHO published Volume 1 of the *WHO monographs on selected medicinal plants*, containing 28 monographs, in 1999, Volume 2 including 30 monographs in 2002, and the third volume presenting 31 monographs in 2007. This fourth volume contains an additional collection of 28 monographs describing the quality control and use of selected medicinal plants.

Each monograph contains two parts, the first of which provides pharmacopoeial summaries for quality assurance purposes, including botanical features, identity tests, purity requirements, chemical assays and major chemical constituents. The second part, drawing on an extensive review of scientific research, describes the clinical applications of the plant material, with detailed pharmacological information and sections on contraindications, warnings, precautions, adverse reactions and dosage. Also included are six cumulative indexes to the four volumes, listing the monographs, the medicinal plant names, and the major chemical constituents.

The *WHO monographs on selected medicinal plants* aim to provide scientific information on the safety, efficacy, and quality control of widely used medicinal plants; provide models to assist Member States in developing their own monographs or formularies for these and other herbal medicines; and facilitate information exchange among Member States. WHO monographs, however, are not pharmacopoeial monographs, rather they are comprehensive scientific references for drug regulatory authorities, physicians, traditional health practitioners, pharmacists, manufacturers, research scientists and the general public.

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